DESIGN AND IN-VITRO CHARACTERIZATION OF METADOXINE BUCCAL PATCHES USING *Borassus flabellifer* FRUIT RESIN - A NOVEL MUCOADHESIVE POLYMER

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Certificate

This is to certify that the **M. Pharm dissertation** entitled **"DESIGN AND *IN-VITRO* CHARACTERIZATION OF METADOXINE BUCCAL PATCHES USING *Borassus flabellifer* FRUIT RESIN - A NOVEL MUCOADHESIVE POLYMER"** being submitted to The Tamil Nadu Dr. M.G.R Medical University, Chennai was carried out by Register number: **261510151** in the **Department of Pharmaceutics**, College of Pharmacy, Sri Ramakrishna Institute of Paramedical Sciences, Coimbatore, under my direct supervision and guidance, to my fullest satisfaction.

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CONTENTS

LIST OF ABBREVIATIONS

ADHD	- Attention Deficit Hyperactivity Disorder
AUC	- Area Under the Curve
AUMC	- Area Under the first Moment Curve
B. flabellifer	- *Borassus flabellifer* L.
BFR	- *Borassus flabellifer* Fruit Resin
C_{max}	- maximum Concentration
cm	- centimeter
et al	- and others
g	- gram(s)
h	- hour(s)
K_{el}	- Elimination rate constant
min(s)	- minutes
mg	- milligrams
mm	- millimeters
ml	- milliliters
MRT	- Mean Residence Time
N	- Newton(s)
nm	- nanometer

PBS	- Phosphate Buffer Solution
PVA	- Polyvinyl alcohol
SA	- Sodium alginate
t_{max}	- time corresponding to C_{max}
$t_{1/2}$	- half life
w/w	- weight/weight
µg	- micrograms
λ_{max}	- absorption maxima

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INTRODUCTION

Tablets have always been the most preferred formulation for drug administration via oral route. Tablets constitute around 70-80% of the total formulations available in the market. However, there are limitations which make tablets as a secondary option when formulating new drugs. This is attributed to the physico-chemical properties as well as pharmacokinetic parameters of the drug intended for formulation such as aqueous solubility, bioavailability, absorption rate and half-life etc. Such limitations can be overcome by opting alternate routes of drug administration. One such route is buccal drug delivery.

Buccal Drug Delivery [1]

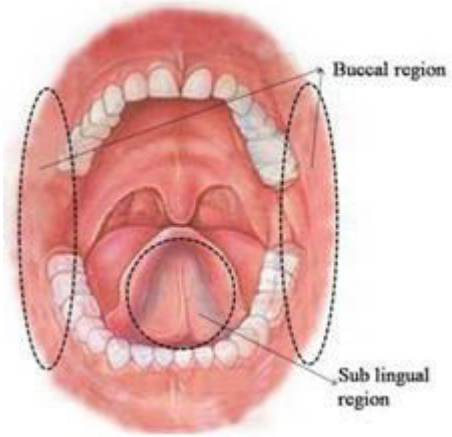


Fig-1: Oral cavity and location of buccal mucosa

Buccal drug delivery is a newly adapted route of drug administration through the mucous membrane, lining the cheeks internally. Buccal drug delivery often involves a formulation which contains bio-adhesive or mucoadhesive material, which adheres to the buccal mucosa over a period of time and releases the drug. Both local and systemic drug action is possible by buccal route. There are two permeation pathways by which the drug gets transferred from the site of adhesion to systemic circulation. They are paracellular (between the cells) and transcellular (across the cells) pathways. The permeating drug can adapt both the pathways simultaneously, but often through one pathway preferably than the other, depending on the physico-chemical properties of the drug. The permeated drug gets absorbed into the reticulated vein which lies underneath the oral mucosa and transported through the facial veins, internal jugular vein, brachiocephalic vein and then drained into the systemic circulation.

A. Structure of buccal mucosa

Buccal mucosa is composed of an outermost layer of stratified squamous epithelium (Figure 2). Below this, lies a basement membrane, a lamina propria followed by the submucosa as the innermost layer. The epithelium is similar to stratified squamous epithelia found in the rest of the body in that it has a mitotically active basal cell layer, advancing through a number of differentiating intermediate layers to the superficial layers, where cells are shed from the surface of the epithelium. The epithelium of the buccal mucosa is about 40-50 cell layers thick, while that of the sublingual epithelium contains somewhat fewer. The epithelial cells increase in size and become flatter as they travel from the basal layers to the superficial layers.

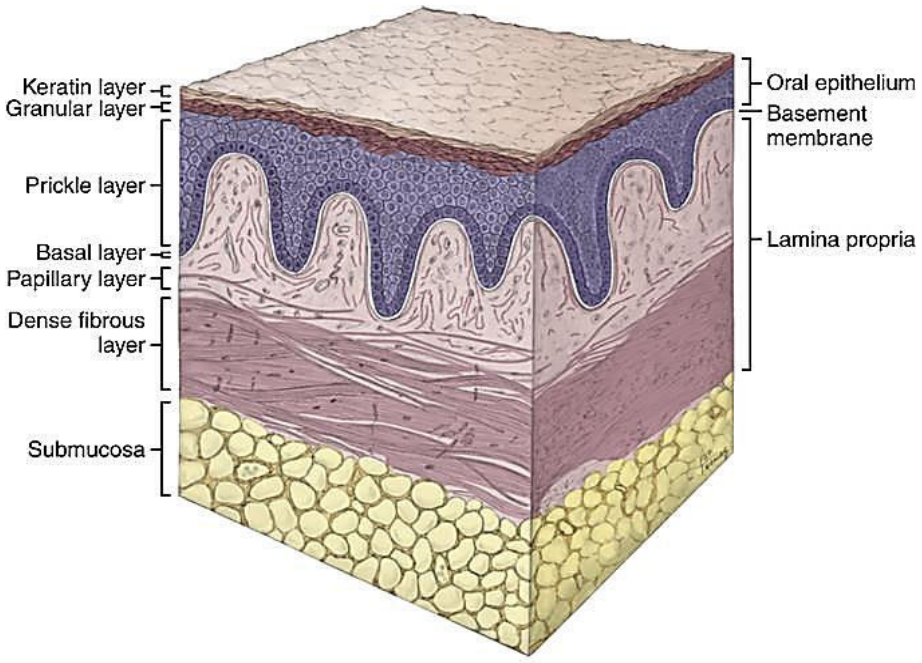


Fig-2: Histology of buccal mucosa

B. Factors affecting absorption of drugs through buccal mucosa

The major factors which affect the absorption of drug through buccal mucosa can be grouped as,

i) Physiological factors - permeability of buccal mucosa

ii) Physico-chemical factors – properties of the drug

i) Permeability of buccal mucosa [1]

The buccal mucosae in general is relatively a leaky epithelial intermediate between that of the epidermis and intestinal mucosa. It is estimated that the permeability of the buccal mucosa is 4-4000 times greater than that of the skin. As indicative by the wide range in this reported value, there are considerable differences in permeability between different regions of the oral cavity because of the diverse structures and functions of the different oral mucosae. In general, the permeabilities of the oral mucosae decrease in the order of sublingual greater than buccal, and buccal greater than palatal. This rank order is based on the relative thickness and degree of keratinization of these tissues, with the sublingual mucosa being relatively thin and non-keratinized, the buccal mucosa is thicker and non-keratinized, and the palatal is intermediate in thickness but keratinized.

ii) Physicochemical properties of the drug [2]

The physicochemical properties which influence the drug absorption through buccal mucosa are described as follows:

a. Molecular weight

Molecules of smaller size penetrate the buccal mucosa better than macromolecules (e.g: peptides) and ions.

b. Degree of ionization

Unionized form of drugs can cross the lipoidal membranes easily compared to their ionized counterparts. Both pK_a of the drug and pH of the buccal environment, which averages from 6.6-6.8, can influence the absorption through buccal mucosa.

c. Partition coefficient

More lipid soluble the compound, higher will be its penetration through the buccal membrane. Hence, compounds of high oil-water partition coefficient (40-200) can permeate well through buccal mucosa.

C. Mechanism of bioadhesion [3]

Bioadhesion may be defined as the state in which two materials, among which, one is of a biological nature, are held together for extend periods of time by interfacial forces. Several theories have been proposed to explain bioadhesion. Any mechanism of adhesion requires the establishment of molecular contact between the bioadhesive material and mucin/epithelial cell surface. In a particular system more than one mechanism may contribute to the formation of bioadhesive bonds which can be specific or non-specific, and can involve covalent or non-covalent bonds. The proposed theories of bioadhesion include,

i. Electronic theory

A double layer of electrical charge is formed at the interface between an adhesive polymer and mucus, due to different electronic characteristics, giving rise to an attractive force from electron transfer across the electrical bilayer.

ii. Adsorption theory

A bioadhesive polymer adheres to mucus, because of a secondary surface force, such as Van der Waal's force, hydrogen bonds or hydrophobic interaction.

iii. Wetting theory

This theory is primarily applicable to liquid bioadhesive systems. Bioadhesion over a wet surface is determined by structural similarity, degree of cross-linking and use of a surfactant.

iv. Diffusion theory

Diffusion theory proposes that polymeric chains of the adhesive and the substrate interacts with each other to a sufficient depth, to create temporary adhesive bond.

The rate of penetration depends upon the diffusion coefficient of the polymer, which in turn is influenced by the molecular weight and cross linking density.

D. Advantages of Buccal drug delivery [4,5]

- Avoiding first pass effect:

Certain drugs undergo extensive first pass metabolism and hence their absolute bioavailability is very less (1-10%). In order to overcome this, drugs are incorporated into a buccal drug delivery system, which exploit the high vasculature of buccal mucosa. Direct access to the systemic circulation through the internal jugular vein avoids acid hydrolysis in the gastrointestinal (GI) tract. Thus this provides an alternative for administration of hormones, narcotic analgesics, steroids, enzymes, cardiovascular agents etc.

- Improved patient compliance in pediatric and geriatric patients:

Children often refuse to ingest tablets or sometimes syrups due to their organoleptic properties. In such cases, buccal drug delivery system may serve as an alternate formulation which may mimic a confectionery. Whereas in elderly people, due to the necessity of taking too many medications (for diabetes, hypertension and hyperlipidemia), they would rather prefer one lesser tablet than usual, whereby a buccal drug delivery system can come in handy.

Termination of the therapy is easy. It can also be administered to unconscious patients.

- Better absorption

The process of absorption via buccal route is passive and rapid. Therefore, it does not require any activation. Among trans-mucosal drug delivery, buccal route lies second to sublingual delivery, while vaginal, rectal, transdermal routes exhibit poor absorption than buccal mucosa.

- Easy to formulate:

The number of intermediate steps involved in formulating a buccal drug delivery system is comparatively very less than a tablet manufacturing process.

E. Disadvantages of Buccal drug delivery

- Drug related issues

Drugs, which irritate the oral mucosa and have a bitter or unpleasant taste and odour, cannot be administered by this route. Drugs, which are unstable at buccal pH cannot be administered by this route.

- Low permeability and surface area:

Buccal mucosa exhibits comparatively low permeability than the sublingual region. The total surface area of the membranes of the oral cavity available for drug absorption is 170 cm^2, of which approximately 50 cm^2 represents non-keratinized tissues, including the buccal membrane

- Salivation:

Salivation can dilute the drug and can make it pass through the pharynx to stomach. Some people possess the nature of excessive salivation wherein the dosage form find difficulties in adhering to the buccal wall.

- Accidental chewing or swallowing:

Children or even adults may accidentally chew off the formulation and the whole purpose of drug delivery through buccal mucosa becomes obsolete. A hazard of choking due swallowing of the patch is also possible.

F. Formulation of buccal drug delivery systems [4]:

The various types of buccal drug delivery systems include buccal tablets, films, patches, gels, ointments and powders. The difference between the term "patch" and "film" may be attributed to their thickness, where the former is thicker than the latter. Buccal patches can be formulated by adopting any of the following methods.

i) Solvent casting method

ii) Direct compression method

iii) Hot-melt extrusion method

i) Solvent casting:

In solvent casting method all the excipients are dispersed in suitable solvents and are mixed together. The mixture is added with required quantity of active pharmaceutical ingredient and allowed to settle till the solution is cleared of entrapped air. Then the solution is poured onto a mold or casting and allowed to dry. Patches of required size and geometry were cut from the parent patch.

ii) Direct compression/milling method:

The drug and the excipients are mixed together as a single physical mixture and kneaded with the help of a minimum quantity solvent. The wet mixture is rolled on the release linear till it achieves desired thickness and allowed to dry.

iii) Hot-melt extrusion method(HME) [5]:

The required polymers are melted during the extrusion process, which can function as thermal binders and act as drug depots upon cooling and solidification. Since solvents and water are not necessary, the number of processing and drying steps are reduced. The intense mixing and agitation imposed by the rotating screw cause de-aggregation of suspended particles in the molten polymer resulting in a more uniform dispersion and the process is continuous and efficient. Bioavailability of the drug substance may be improved when it is solubilized or dispersed at the molecular level in HME dosage forms. Pharmaceutical Hot-Melt Extrusion processes can be categorized as either ram extrusion or screw extrusion.

Though there are no significant differences in the performance of the patches prepared by the above methods, solvent casting method is least preferred due to the possibility of residual solvent in the formulation and the corresponding solvent related health issues.

G. Composition of buccal patches [6]

a) Active pharmaceutical ingredient:

The selection of drug is a vital process when formulating a buccal patch. Potent drugs, i.e., drugs with a conventional dose of lesser than 100mg and drugs which undergo extensive first pass metabolism (which also exhibit low bioavailability, less than 20%) are preferred candidates for buccal drug delivery. Buccal patches are generally intended for sustained release, hence drugs with a half-life of 2-8 hours will be an apt candidate. pH is another factor to be considered when choosing a drug, since the drug or even any excipient should not irritate the buccal mucosa, due to acidity or alkalinity.

b) Polymers:

To serve as mucoadhesive polymers, the polymers should possess some general physiochemical features such as,

- ✓ Predominantly anionic hydrophilicity with numerous hydrogen bond-forming groups.
- ✓ Polymer and its degradation products should be non-toxic, non-irritant and free from leachable impurities.
- ✓ Should have good spreadability, wetting, swelling and solubility and biodegradability properties.
- ✓ pH should be biocompatible and should possess good viscoelastic properties.
- ✓ Should possess peel, tensile and shear strengths at the bioadhesive range.
- ✓ Should possess bioadhesive, film-forming and if required, sustained release properties.

Such polymers include Hydroxy propyl cellulose(HPMC), Hydroxy ethyl cellulose(HEC), Polyvinyl alcohol(PVA), Carbopol, Polyvinylpyrrolidone (PVP) etc.

c) Diluents:

If the drug is potent (dose less than 10mg), a suitable diluent such as Lactose may be added to increase the bulk of the formulation. Diluent such as microcrystalline cellulose is used when buccal tablets or patches are formulated by direct compression method.

d) Sweetening agent:

Sucralose, Aspartame, Mannitol etc., can be used as sweetening agents.

e) Flavouring agent:

Vanillin, Clove oil, Menthol etc., may serve as suitable flavouring agents.

f) Plasticizer:

Plasticizer is a vital ingredient which determine most of the physical properties of the buccal patch such as elasticity, folding endurance, tensile strength etc. Hence reliable plasticizers such as Polyethylene glycol 100, 400, Propylene glycol and Dibutyl phthalate can be used.

g) Permeation enhancer:

Examples of permeation enhancers include Dimethyl sulphoxide (DMSO), Sodium taurocholate, Sodium glycocholate, Oleic acid, Cyanoacrylate etc.

H. Evaluation of buccal patches [7]

Buccal patches are mainly evaluated for their physical properties, bioadhesion and release properties. The following are the important evaluation parameters with regard to buccal patches:

a) Thickness:

Thickness determines the uniformity of content in the patch and also its aesthetic value to an extent. Thickness can be measured using a digital screw gauge or calibrated digital micrometer.

b) Weight variation:

Weight variation is also determined to ensure content uniformity, since deviation in uniform weight is due to difference in the amount of either drug or the polymer matrix in an individual patch.

c) Folding endurance:

Folding endurance is determined by repeated folding of the patch at the same place till it breaks. The number of times the patch is folded without breaking is recorded as the folding endurance value. The concentration of plasticizer is responsible for the folding endurance of a patch

d) Tensile strength:

Tensile strength is the maximum stress applied to a point at which the buccal patch breaks. Tensile strength can be calculated by the equation.

$$\text{Tensile strength}(kg/mm^2) = \frac{\text{Force at break}}{\text{Initial cross-sectional area}}$$

e) Drug content:

Determination of drug content in buccal patches can be carried out by, suitably dissolving the patches and diluting, to a get a clear solution, which can be estimated by any of the analytical methods such as UV spectrophotometry, fluorimetry, HPTLC or HPLC as suggested in the monograph of the drug incorporated. A sample of 3 patches may be subjected to the assay procedure to ensure the content uniformity.

f) Swelling index:

Swelling index is determined using simulated saliva solution (pH 6.8 buffer solution). Each patch is weighed and placed in a pre-weighed stainless steel wire mesh. The mesh containing the patch is submerged into 4ml medium. Increase in the weight of the film was determined at pre-set time intervals until a constant weight is seen. The degree of swelling can be calculated using the following equation:

$$\text{Swelling index} = \frac{\text{Weight after swelling} - \text{Initial weight}}{\text{Initial weight}}$$

g) Surface pH:

Determination of surface pH of a buccal patch formulation ascertains that the patch does not cause any local irritancy to the buccal mucosa. A number of methods can be employed to determine surface pH such as use of pH paper or digital pH meter, over the surface of the formulation, previously wetted with water.

h) In-vitro bioadhesion [8]:

Several techniques such as tensile strength testing, adhesion weight method, fluorescent probe method, flow channel technique, colloidal gold staining method are employed to determine bioadhesive strength of buccal patches.

A fabricated setup which consist of a modified physical balance is used to determine the bioadhesive strength of the buccal patches.

i) In-vitro diffusion/permeation [9]:

A Franz diffusion cell can be used to study the drug diffusion or permeation pattern. Cellulose nitrate filter can be used as an artificial membrane to mimic the buccal mucosa [10]. The diffusion medium used is phosphate buffer solution (PBS) at a pH of 6.8, which acts as simulated saliva.

j) In-vivo bioavailability [11]:

In-vivo bioavailability studies are preferably carried out with the help of human volunteers. Proper permission must be sought from the ethical authorities, prior to the study. An informed consent must be given to the volunteers and the Declaration of Helsinki guidelines must be followed throughout the course of the study.

The buccal patches must be administered to the volunteers and blood samples are collected periodically. The samples are subjected to suitable extraction methods and analyzed for the concentration. The pharmacokinetic parameters such as C_{max}, t_{max}, K_{el}, $t_{1/2}$, AUC, AUMC and MRT are determined using the plasma concentration-time data and thus bioavailability is calculated.

The other methods specific for monitoring bioadhesion include Gamma scintigraphy, isolated loop technique and transit studies with radiolabelled or fluorescent coupled dosage forms.

REVIEW OF LITERATURE

Choy Fun Wong *et al*., **(1999)** [12], fabricated controlled release buccal patches containing Metoprolol tartrate, using Eudragit-NE40D, along with other polymers such as HPMC, Sodium CMC and Carbopol of different grades to modify the mucoadhesive property. Although high viscosity polymers can enhance the bio-adhesiveness of the patches, they also tend to cause non-homogeneous distribution of the polymers and drug, resulting in non-predictable drug-release rates. Of the various bioadhesive polymers studied, Cekol-700 appeared to be most satisfactory in terms of modifying the drug release and enhancement of the bioadhesive properties.

Addolorato. G *et al*, **(2003)** [13], have reviewed about the use of Metadoxine in the treatment of acute and chronic alcoholism. In this review, the preclinical and clinical results obtained using Metadoxine in acute and chronic alcohol intoxication were reported. Metadoxine also seems to be safe; in more than 15 years of post-marketing surveillance. Only minor aspecific and reversible events were observed in patients exposed to the treatment.

Neeraj Kaul *et al*., **(2005)** [14], have developed a stability indicating method for determination of Metadoxine in pharmaceutical dosage forms, using HPTLC. The method involved use of TLC Aluminium plates pre-coated with silica gel 60F-254 as the stationary phase. The solvent system consisted of acetone–chloroform–methanol–ammonia (7.0: 4.0: 3.0: 1.2, v/v/v/v). Densitometric analysis of Metadoxine was carried out in the absorbance mode at 315 nm. Metadoxine was subjected to acid, alkali and neutral hydrolysis, oxidation, dry and wet heat treatment and photo and UV degradation. The drug undergoes degradation under all stress conditions. Also, the degraded products were well resolved from the pure drug with significantly different R_f values.

Pradeep kumar *et al*., **(2008)** [15], have developed and validated a spectrophotometric method to estimate Metadoxine in bulk and pharmaceutical dosage forms using derivative spectroscopy. The method was based on Metadoxine, showing absorbance at 292, 302, 270 and 314 nm for zero order, first order, second order and third order derivative spectroscopy respectively in distilled water. But regression values with best results were found to be best for third order derivative spectroscopy. The method obeyed Beer's law in the concentration range of 4 to 40µg/ml. The proposed method was precise, accurate, linear, stable and reproducible and can be extended to the analysis of Metadoxine in bulk and tablet formulations.

Surya N. Ratha *et al*., **(2010)** [8], have attempted to formulate a buccal drug delivery system containing Atenolol using Sodium alginate along with various hydrophilic and mucoadhesive polymers like CP 934 P, Sodium CMC, and HPMC in various proportions and combinations. Buccal patches of Atenolol provided sustained buccal delivery of atenolol for a long period and promised to be a good way to bypass the extensive hepatic first-pass metabolism in the management of hypertension.

P. Chinna Reddy *et al*., **(2011)** [5], have reviewed on bioadhesive buccal drug delivery systems and their current status of formulation and evaluation methods. This article describes about the nature of buccal mucosa and pathways of drug absorption through it; it's advantages, limitations and scope for improvisation. They have also elaborated on the various types of buccal drug delivery systems, formulation techniques, composition of buccal drug delivery systems and their evaluation techniques.

Ikoni J. Ogaji *et al*., **(2011)** [16], have reviewed the current applications of natural polymeric materials in pharmaceutical formulations. The pharmaceutical applications of some of the traditional and commercially available natural polymers were discussed. Emerging potential pharmaceutical excipients of natural origins such as Xyloglucan, Pullulan, Pectin, starches, *Moringa oleifera* gum etc., were also discussed.

Ravi Kumar *et al*., **(2012)** [17], have isolated and characterized mucilage obtained from unripened fruits (endosperms) of *Borassus flabellifer*. This study elucidated the physical, thermal, sorption and functional properties of *Borassus flabellifer* mucilage, by elemental analysis, Fourier transmittance infra-red analysis, particle size analysis, thermogravimetric analysis, differential scanning calorimetry, scanning electron microscopy and X-ray powder diffraction.

In a different study, **Ravi Kumar** *et al*., **(2012)** [18], have also studied the use of mucilage obtained from fruits of Borassus flabellifer as a natural gelling agent. Mucilage extracted from endosperm of *Borassus flabellifer* fruit was subjected to toxicity studies for its safety and preformulation studies for its suitability as a gelling agent. Diclofenac sodium was used as model drug for the formulation of gels. Thus it was concluded that the *Borassus flabellifer* mucilage can be used as a pharmaceutical excipient in gel formulations and it has the potential to replace some synthetic gelling polymers upon further modifications.

Waleed Khattab *et al*., **(2013)** [19], have formulated buccal tablets containing Zolmitriptan using different mucoadhesive polymers (natural and synthetic) in different ratios by direct compression technique. All tablets showed acceptable mucoadhesive strength. In-vitro drug release studies showed that at least 76 % of the drug was released within 7 hrs. Release of Zolmitriptan from all tablets followed zero order kinetics. Hence, these formulations of are promising ones as a controlled drug delivery system that will lead to improved bioavailability and greater therapeutic efficacy.

Amelia M. Avachat *et al*., **(2013)** [20], developed mucoadhesive buccal films using tamarind seed xyloglucan (TSX) as novel mucoadhesive polysaccharide polymer for systemic delivery of Rizatriptan benzoate through buccal route. Ex vivo diffusion studies were carried out using Franz diffusion cell, while bioadhesive properties were evaluated using texture analyzer with porcine buccal mucosa as model tissue. This study suggests that tamarind seed polysaccharide can act as a potential mucoadhesive polymer for buccal delivery of a highly soluble drug like Rizatriptan benzoate.

Khadir. A *et al*., **(2013)** [10], have investigated the use of model polymeric membranes; cellulose acetate and cellulose acetate-nitrate, as an alternative to the natural mucosa. Permeability coefficient and steady-state flux for Carvedilol were determined in natural and artificial membranes. The effect of chemical enhancers on permeability through polymeric membranes was measured and compared against that in porcine and rabbit mucosa. A strong and statistically significant correlation between artificial membranes and buccal mucosa for the delivery of carvedilol was established.

Ann Rose Augusthy *et al*., **(2014)** [21], have attempted to formulate buccal patches containing Rabeprazole sodium using HPMC, PVP and Gelatin. The patches were prepared and evaluated for their thickness uniformity, folding endurance, weight uniformity, content uniformity, and in-vitro release studies were conducted for Rabeprazole loaded patches in phosphate buffer (pH-6.8) solution.

Jeevan Sen *et al*., **(2014)** [22], have studied the in-situ gel forming properties of Chitosan and Gellan gum, in the administration of Clotrimazole as a vaginal mucoadhesive drug delivery. Conventional topical application of Clotrimazole to skin may cause localized irritation of the skin with a mild burning sensation, redness and itching. The formulations were characterized for various in-vitro parameters e.g. clarity, pH, isotonicity, viscosity, drug release profile, statistical release kinetics, bioadhesive force, retention time, microbial efficacy, irritation test and stability studies. The developed formulation was found to be non-irritant, bioadhesive with good retention properties. Hence the developed formulation was suggested as an alternative for vaginal dosage forms.

Shivhare. U.D *et al*., **(2014)** [9], have formulated buccal patches loaded with Aceclofenac using HPMC-E15 and Eudragit RL-100, in an attempt to enhance the bioavailability of the drug, which is usually 40-50%, due to extensive first pass metabolism. This study was also focused reducing the dosing frequency of the drug by formulating it into a sustained-release form. Among five formulations, patch prepared with 1:1 ratio of HPMC-E15 & Eudragit RL-100 showed maximum release 92.35% up to 8 hours.

Bhattacharjee. S et al., **(2014)** [23], have studied the effect of varying concentrations of plasticizer on the physical properties of mucoadhesive films. Buccal films were prepared by using Hydroxy Propyl Methyl Cellulose (HPMC) as the polymer and Glycerine, Propylene glycol, Dibutyl phthalate, Triethanolamine as plasticizers. Methanol and Acetone were used as solvents. It was concluded that buccal films prepared by using Propylene glycol as the plasticizer in the solvent Methanol, promotes sustained drug release over a period of 6 hours of study and hence proved to be a good plasticizer in formulating buccal films.

Vengaiah. P.C et al., **(2015)** [24], have studied *Borassus flabellifer* fruit pulp for its physico-chemical properties. From this study, it was observed that the fresh pulp contains a moisture content of 74.5%. The ash and fat contents (wet matter basis) were 1.2% and 0.8% respectively. The protein content and carbohydrate content were 1.25% and 22.5% respectively. The calorific value obtained was 102.83 kcal/100g. The pH value was 5.5. Water absorption capacity (18%) and bulk density (0.78 g/cm3) were recorded for the Palmyra fruit pulp. The values for swelling power and foam capacity were 4% and 2.5% respectively.

Saranya. P and **Poongodi Vijayakumar. T**, **(2016)** [25], have carried out a phytochemical screening of raw and thermally processed *Borassus flabellifer* fruit pulp. The results revealed that both the raw and processed Palmyra palm fruit pulp contain all the phytochemicals such as carbohydrates, alkaloids, flavonoids, tannins, glycosides, saponins and phenolics, except proteins. On processing, alkaloids were reduced in the aqueous extract of processed pulp and saponins in both the extracts of raw and processed pulp. Therefore, no major changes in the presence of phytochemical components due to heat processing were found in this current study.

SCOPE OF THE WORK

Buccal drug delivery seems to be a boon for formulation researchers, especially when formulating a sustained release formulation or when facing a difficulty with a drug which undergoes first-pass effect. The polymers used are also of natural or semisynthetic origin, which gives a wider scope of using natural polymers. This work focuses on using a novel mucoadhesive polymer obtained from Palmyra palm.

1. Palmyra palm fruit resin-a novel mucoadhesive polymer:

A number of researchers have worked on using polymers of natural origin as excipients in novel drug delivery systems, since natural polymers offer the advantage of biocompatibility and abundance. One such polymer of plant origin which has been underutilized in spite of its numerous uses is resin obtained from the fruit pulp of Palmyra palm.

Palmyra palm or Asian palm is a tall erect tree abundantly found in Asian countries from the Persian Gulf to the Cambodian-Vietnamese border and are specifically cultivated in India, Sri Lanka, Malaysia and in some American regions like Hawaii and Florida.

Fig-3: Palmyra palm trees

Each palm may bear 6-12 bunches of about 50 fruits per year. An average crop of Palmyra palm in Sri Lanka is 350 fruits. In India, it is grown as a windbreak over the plains. The coconut-like fruits are three-sided when young, 12-15 cm wide, and capped at the base with overlapping sepals. The outer covering is smooth, thin, leathery and brown, turning nearly black after harvest. Inside is a juicy mass of long, tough, coarse, white fibers coated with yellow or orange pulp. When the fruit is very young, this kernel is hollow, soft as jelly, and translucent like ice and is accompanied by a watery liquid, which is sweetish and potable. The pulp of mature fruits is sucked directly from the wiry fibers of roasted, peeled fruits. It is also extracted to prepare a product called **punatoo** in Sri Lanka. It is eaten alone or with the starch from the Palmyra seedlings. The fresh pulp is reported to be rich in vitamins A and C. The pulp of the mature fruit can be used in the treatment of dermatitis [26].

This processed fruit pulp is usually a sticky mass and remains in the mouth for a longer period. This formed the basis for using this resinous extract as mucoadhesive polymer.

2. Alcoholism

People of the modern world assume social drinking (alcohol consumption) as an indication of one's social status or the ability to get along with the society or peer group. In the course of this action, they often tend to forget the serious adverse effects associated with alcohol consumption, especially the level of damage that one's liver might be subjected to. Alcohol intoxication can be described as a change or a disturbance in the behaviour or mental function of an individual during or after consumption of alcohol.

More excessive or serious forms of drinking, often chronic, may be potentially harmful to the drinker and to others affected by the drinker. For example, drinking with the intent to get drunk or intoxicated, drinking and driving while under the influence of alcohol, loss of psychomotor coordination and speech, blackouts,

vomiting and alcohol poisoning are considered among the harmful symptoms and effects of drinking that falls outside the scope of social drinking. In addition to the effects of alcohol on the nervous system, it may be one of the major contributing factors to road accidents, suicide and violent death in young adults. Alcohol intoxication can affect a person's judgement. Sometimes, coma can occur. Alcohol intoxication is managed with rest, hydration and stopping alcohol use. Severe cases require hospital admission, intravenous fluids, observation and supportive care. Alcohol abuse, moreover, is a common problem in the general population all over the world. Alcohol abuse and alcoholism are responsible for a wide variety of medical problems, which are considered part of the new-age epidemics, among them the most recognized being alcohol-induced liver disease, primary and secondary malnutrition, and neuron damage, often leading to death. It would be desirable to avoid these and other effects or consequences of excessive alcohol consumption.

In the Indian scenario, more than 10 million cases of alcohol intoxicated patients are reported per year (sources: Apollo Hospitals). As the debate over alcohol ban grows across India, 15 people die every day – or one every 96 minutes – from the effects of drinking alcohol, reveals an India Spend analysis of 2013 National Crime Records Bureau (NCRB) data, the latest available. The per capita consumption of alcohol in India increased 38%, from 1.6 litres in 2003-05 to 2.2 litres in 2010-12, according to a World Health Organization (WHO) report, which also revealed that more than 11% of Indians were binge drinkers, against the global average of 16 percent. Before the latest crackdowns on alcohol, Gujarat and Nagaland were the only Indian states with prohibition. Maharashtra tops alcohol-related deaths. Maharashtra reported the most alcohol-related deaths, followed by Madhya Pradesh and Tamil Nadu, according to the NCRB data, with experts saying high rates of alcoholism correlate with high crime rates [27].

The most lasting damaging actions of ethanol are exerted on the liver function and structure. A liver disease is often present in patients affected by alcoholism; however, the mechanisms responsible for the liver toxicity of ethanol are still not completely understood. Ethanol also modifies the GABA-mediated neurotransmission. It preferentially stimulates the dopaminergic transmission in the mesolimbic system, interferes with serotoninergic transmission and with the release of glutamate in the central synapses. The neuropathological manifestations usually appear after many years of excessive drinking. The pharmaco-therapeutic aspect of alcoholism includes the use of drugs, with different actions and objectives. Among them, **Metadoxine** seems to be of interest [13].

Therefore, Metadoxine was chosen as a model drug for the development of buccal patches as an attempt to improve compliance in chronic alcohol addicts.

3. What's new?

I. Usually films or patches, either transdermal or buccal involves a dose calculation based on the surface area. In this study, the 'thickness' factor is incorporated, enabling a more precise dose calculation, since the volume of the matrix is considered, i.e., a patch is considered as a three dimensional cylinder rather than a two dimensional circle. The dose calculation is proceeded as follows:

Volume of a parent patch made up by a particular volume of polymer mixture/matrix $= \pi R^2 h$

Volume of individual patch (final product) $= \pi r^2 h$

where R = radius of parent patch

r = radius of individual patch

The number of possible patches (theoretically) $= \frac{\text{Volume of parent patch}}{\text{Volume of individual patch}}$

Thus, the quantity of drug to be added = Number of theoretical patches × Dose of individual patch

II. A new laboratory scale hand punch for cutting the patches from a parent patch was fabricated, which helps in cutting the patches of fixed and uniform diameter. The base and the punches were fabricated with stainless steel. The punches with sharp edges, are screwed onto the base, so as to facilitate addition of new punches of variable diameter.

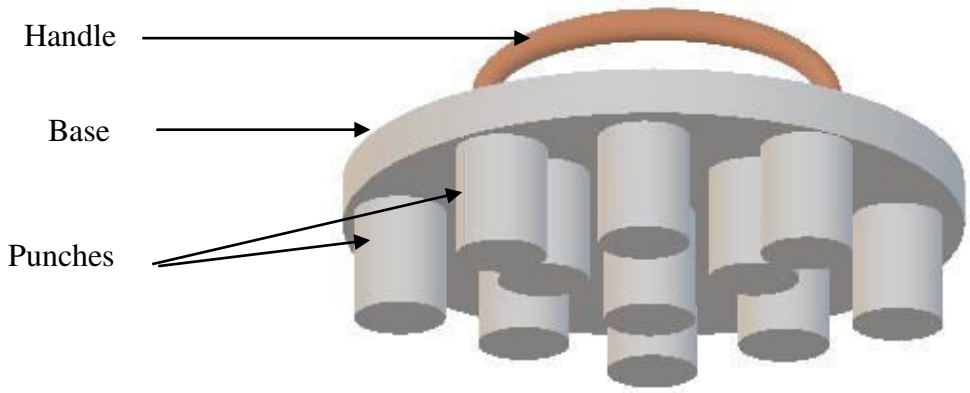


Fig-4: Fabricated Patch Cutter

DRUG PROFILE

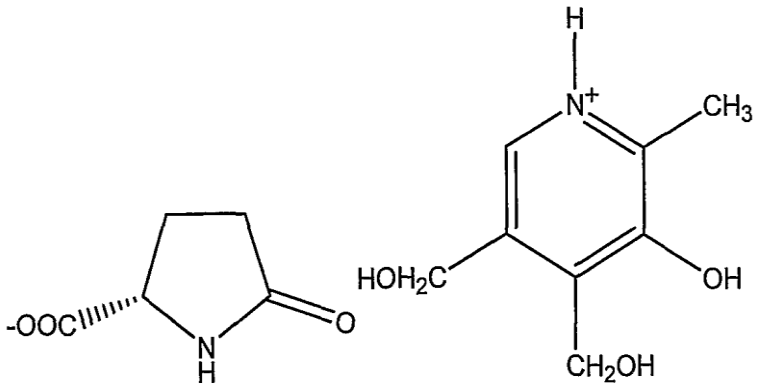
METADOXINE [28]

Synonyms : pyridoxine L-2-pyrrolidone-5-carboxylate

IUPAC name : 4,5-bis(hydroxymethyl) - 2- methylpyridin – 3 -ol;
(2S) – 5 – oxopyrrolidone – 2 - carboxylic acid

Empirical formula : $C_{13}H_{18}N_2O_6$

Chemical structure :



Molecular weight : 298.295 g/mol

Appearance : white or almost white crystalline powder

Solubility : freely soluble in water and methanol, soluble in Ethanol, insoluble in chloroform and Diethyl ether

Melting point : 97°-100° C, meanwhile decomposition on melting is observed

Optical rotation : -9.0° to -12.5°

Category : Hepatoprotective

Brand names : Metadoxil (Micro Labs Ltd.), Alcoliv (Sun Pharmaceutical Industries Ltd.), Viboliv (Dr. Reddy's Laboratories Ltd.), Livodox (Icon Life Sciences Ltd.), Toneliv (Esmatrix Life Sciences Ltd.)

Mechanism of action [29]:

Metadoxine is a selective antagonist of the serotonin receptor subtype 5-HT_{2B} and displays high affinity to the gamma-aminobutyric acid (GABA) transporter. *In vitro* enzymatic assay revealed that Metadoxine reduced the activity of the GABA transaminase enzyme, responsible for the degradation of GABA. Electrophysiological studies also showed that Metadoxine increased inhibitory GABA based synaptic transmission via a presynaptic effect. As it does not affect dopamine, norepinephrine or serotonin levels, Metadoxine displays a novel mechanism of action as a monoamine-independent GABA modulator.

Its primary effect is to increase elimination of alcohol via the kidneys, and to help clear the by-products of alcohol decomposition, such as acetaldehyde, from the blood and tissues. The process of oxidizing ethanol into acetaldehyde and acetone consumes reduced glutathione levels. Following a consumption of alcohol, Metadoxine helps restore nicotinamide-adenine-dinucleotide (NAD), glutathione, and adenosine triphosphate (ATP) concentration in the liver and the brain, as well as normalizes alanine aminotransferase (ALT), aspartate aminotransferase (AST), and gamma-glutamyl transpeptidase (GGT) levels, all of which are characteristic signs of liver regeneration.

Pharmacokinetics [13]:

- The oral absorption of the drug is fast, with high and reproductive absolute bioavailability (60 to 80%).
- It undergoes extensive tissue distribution and hence large apparent distribution volume is observed.
- The half-life is 40 to 60 minutes without appreciable differences between oral or intravenous administration.
- Excretion occurs approximately in the same proportion through the urine and the feces, between 40 and 45% in 24 hours in the urine, and between 35 and 50% in 96 hours in the feces.

Indications:

- Treatment of alcohol intoxication; 500-1000mg orally twice daily
- Treatment of fatty liver - both alcoholic and non-alcoholic
- Treatment of ADHD
- Treatment of Fragile X Syndrome

Side effects [30]:

No unfavorable side effects are reported that could be attributed to the drug. Therefore, Metadoxine be considered a valuable resource in the treatment of **alcoholic liver disease**.

POLYMER PROFILES

I. *Borassus flabellifer* Fruit Resin

Synonyms : Asian palm, Palmyra palm, Toddy palm, Cambodian palm

Source : Ripened fruit pulp of *B. flabellifer*

Taxonomy [31,32] :

Kingdom	Plantae
Subkingdom	Viridiplantae
Infrakingdom	Streptophyta
Superdivision	Embryophyta
Division	Tracheophyta
Subdivision	Spermatophytina
Class	Magnoliopsida
Superorder	Lilianae
Order	Arecales
Family	Arecaceae
Genus	*Borassus* L.
Species	*Borassus flabellifer* L.

Appearance : Golden brown or dark brown sticky resinous substance

Odour : Sweet and fruity smell

Solubility : Soluble in water, sparingly soluble in Methanol, insoluble in Chloroform and Dichloromethane.

Constituents [25] : Hemicellulose is the primary constituent of this resin, while it also contains traces of phyto-constituents like alkaloids, flavonoids, saponins, tannins & vitamins.

pH : 5.5 – 6

Swelling ratio [24] : 4 g/g

Moisture content : 74-77 %

Water absorption capacity : 3 %

Stability

The resin is thermally unstable (above 60°C). But exhibits remarkable stability on storage at normal conditions (room temperature in an air-tight container). Even after 150 days since extraction, no fungal or bacterial growth was observed.

Applications in pharmaceutical formulations & technology:

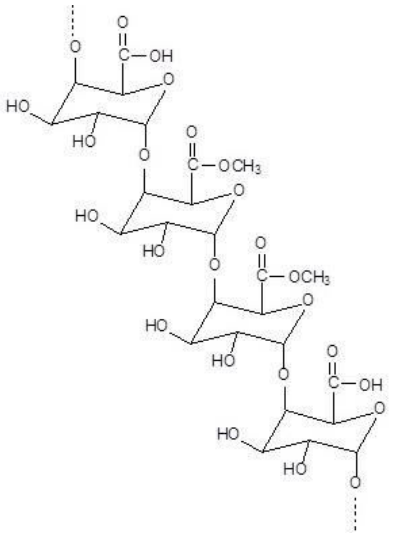
- The resin or the mucilage (prior to drying) has been studied for its gel forming properties, using Diclofenac sodium as a model drug [17].
- It can be opted to replace synthetic or commercially available polymers to be used as a gelling agent or binding agent.
- It also possesses inherent stomachic, sedative, laxative, aphrodisiac and anti-inflammatory properties [25].

II. Pectin [16]

Synonyms : Cellulose, Kaopectate

Source : Pectin is mainly obtained from citrus peel or apple pomades, both of which are by-products of juice manufacturing process Apple pomade contains 10–15% of pectin on a dry matter basis while Citrus peel contains of 20–30%.

Chemical structure :



Molecular weight : 194.139 g/mol

Constituents : Pectin is mainly composed of D-galacturonic acid units joined in chains by means of á-(1-4) glycosidic linkage.

Functional Category : Stabilizing agent; gelling agent; thickening agent

Description : Off-white colored amorphous, odorless, free-flowing, fine powder

pH : 3.2-3.4

Heat of combustion : 14.6 J

Specific gravity : 1.6 at 25°C

Solubility : soluble in ordinary water, partially soluble in cold water, insoluble in organic solvents and alcohol

Application in pharmaceutical formulation or technology

Pectin is widely used in pharmaceutical formulations as gelling agent, thickener, water binder and stabilizer. It is compatible with most of the other pharmaceutical ingredients. It also has good stability and viscosity over a wide pH and temperature ranges.

Safety

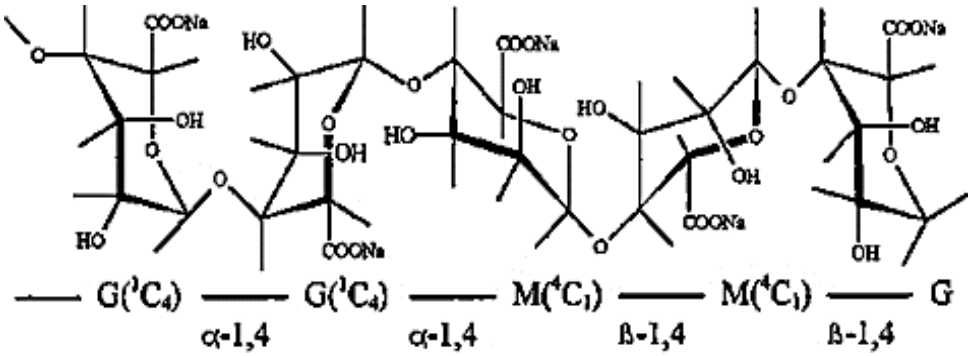
Pectin is non-toxic and non-irritant at the levels employed as a pharmaceutical excipient

III. Sodium alginate [33]

Synonyms : Algin, Sodium salt of Alginic acid, Kelcosol, Keltone, Manucol, Manugel, Pronova, Satialgine-H8

Source : It is extracted from seaweed, algae, and bacteria

Chemical structure :



Molecular weight : 1828 g/mol

Constituents : Alginate is composed of (1–4)-b-D-mannuronic acid (M) and (1–4)-a-L-glucuronic acid (G) units in the form of homo polymeric (MM- or GG-blocks) and hetero polymeric sequences (MG or GM-blocks)

Functional category : Stabilizing agent; suspending agent; tablet and capsule disintegrant; tablet binder; viscosity-modifier.

Description : white to pale yellowish brown colored powder

pH : 7.2

Melting point : 20°C

Specific gravity : 1.26

Viscosity : 20-400 mPa

Solubility : slowly soluble in water, forming a viscous colloidal solution; practically insoluble in Ethanol, Diethyl ether, other organic solvents and acids.

Application in pharmaceutical formulation or technology

Alginate and their derivatives are widely used by many pharmaceutical scientists for drug delivery and tissue engineering applications due to its many properties such as biocompatibility, biodegradability, water solubility, relatively low cost, gelling ability, stabilizing properties, and high viscosity in aqueous solutions. In topical formulations Sodium alginate is used as a thickening and suspending agent in variety of creams and gels and as a stabilizing agent for oil in water emulsion.

Safety

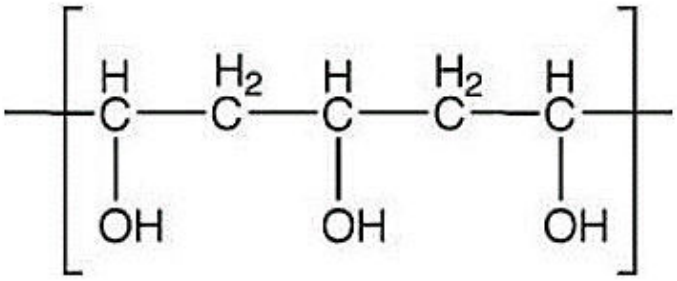
Low toxicity & non-immunogenicity are salient safety features of Sodium alginate, although excessive oral consumption may be harmful.

IV. Polyvinyl Alcohol [34]

Synonyms : Poly(Ethenol), Ethenol, homopolymer, Polyviol, Vinol, Alvyl, Alcotex, Covol, Elvanol, Gelvatol, Lemol, Mowiol

Source : It is synthesized by the polymerization of vinyl acetate to polyvinyl acetate (PVAc) which is then hydrolysed to get PVA.

Chemical structure :



Molecular weight : Ranges from 4000-200000 (the one which is used in this study is 160000), with a repeating monomer of weight 44.00 g/mol

Functional category : film-forming agent; gelling agent; viscosity modifier; surfactant; bioadhesive.

Description : colorless crystalline substance

pH : 5.0-6.5

Melting point : 200°C

Specific gravity : 1.35

Solubility : PVA is soluble in highly polar and hydrophilic solvents, such as water, Dimethyl Sulfoxide(DMSO), Ethylene Glycol (EG), and N-Methyl Pyrrolidone (NMP). The solubility of PVA in water depends on the degree of polymerization (DP), hydrolysis, and solution temperature.

Application in pharmaceutical formulation or technology

PVA hydrogels have been used for various biomedical and pharmaceutical applications. It is bioadhesive in nature. It has high tensile strength and flexibility. PVA also shows a high degree of swelling in water (or biological fluids) and a rubbery and elastic nature and therefore closely simulates natural tissue and can be readily accepted into the body. PVA gels have been used for contact lenses, the lining for artificial hearts, and drug- delivery applications. PVA is mainly used in topical pharmaceutical and ophthalmic formulations. It is used as a stabilizer in emulsions. PVA is used as a viscosity increasing agent for viscous formulations such as ophthalmic products. It is used as a lubricant for contact lens solutions, in sustained release oral formulations and transdermal patches.

Safety:

Advantages of PVA hydrogels are that they are non-toxic and non-carcinogenic

OBJECTIVES OF THE STUDY

- To establish the mucoadhesive property of a natural polymer obtained from fruits of *B. flabellifer* (Palmyra palm) plant.

- To determine whether this novel natural polymer possess sustained release or immediate release pattern.

- To formulate the drug Metadoxine in a novel dosage form, which is currently unavailable in the market.

- To propose a new theory for calculation of dose, by incorporating thickness factor.

- To determine whether the change in dosage form can enhance the bioavailability and thereby reduce the dosing frequency as well as dose dumping of the particular drug.

PLAN OF THE WORK

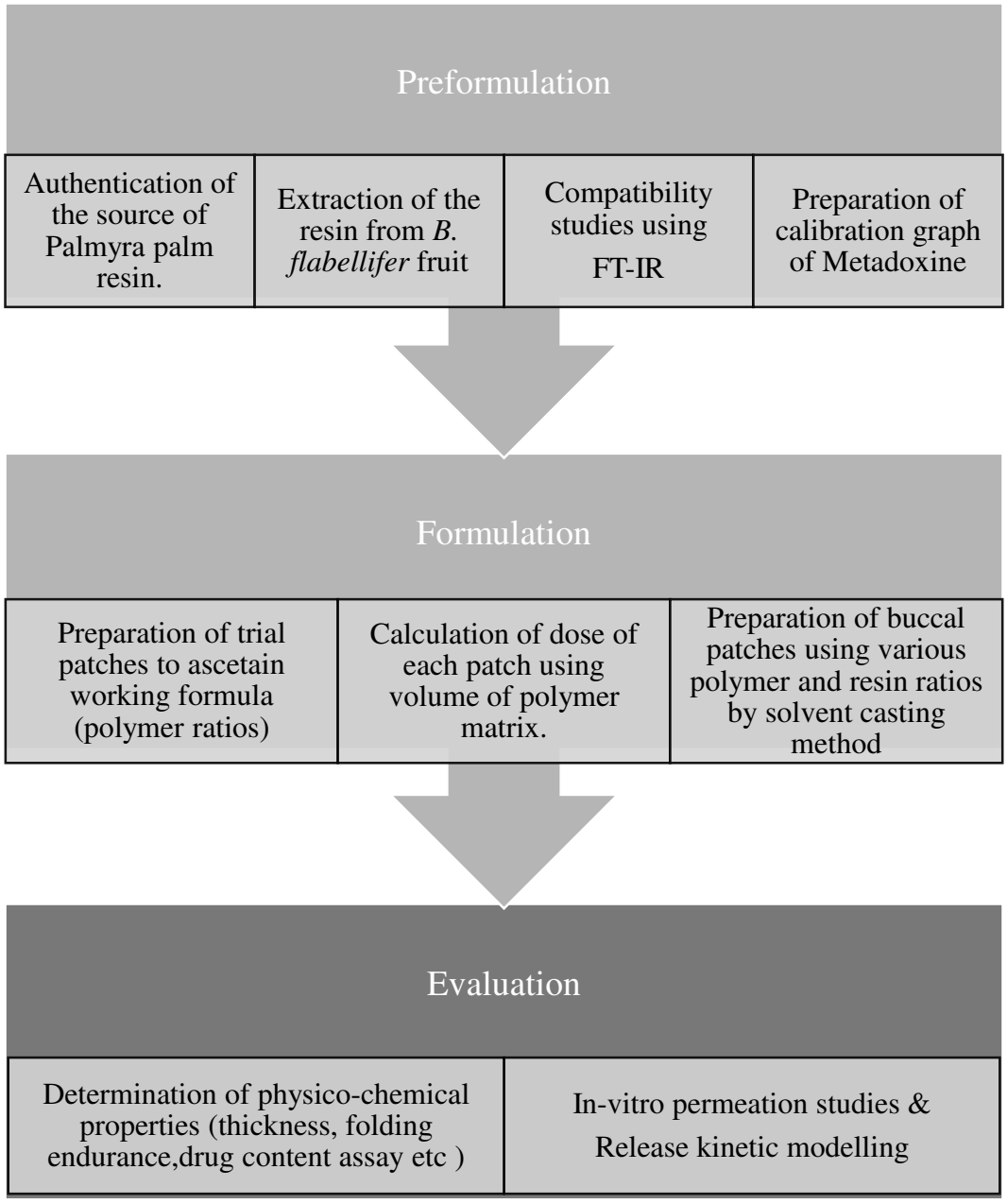


Fig-5: Plan of the work

MATERIALS & EQUIPMENTS

Table-1: List of materials and reagents used in the study

Reagent	Manufacturer
Borassus flabellifer Resin	Natural source
Pectin	Himedia Laboratories, Mumbai
Sodium alginate	S.D Fine Chemicals Ltd., Mumbai
Polyvinyl alcohol (M.W:160000)	Himedia Laboratories, Mumbai
Sucrose	S.D Fine Chemicals Ltd., Mumbai
Vanillin	Himedia Laboratories, Mumbai
Metadoxine	Apotex Research Pvt. Ltd.
Polyethylene glycol - 400	Himedia Laboratories, Mumbai
Dimethyl sulphoxide	E. Merck Ltd., Mumbai
Potassium dihydrogen phosphate	S.D Fine Chemicals Ltd., Mumbai
Sodium hydroxide	S.D Fine Chemicals Ltd., Mumbai
Potassium Bromide	Himedia Laboratories, Mumbai
Cellulose nitrate filter	Sartorius

Table-2: List of equipment and apparatus used in the study

Equipments	Model/Company
UV/ Visible Spectrophotometer	JASCO V-630
Franz Diffusion Cell	Fabricated
FT-IR Spectrometer	JASCO-4100
Magnetic stirrer	Remi Equipment
Hot air oven	INLAB Equipments Pvt. Ltd.
Electronic digital micrometer	Aerospace, China
Electronic balance	Shimadzu electronic balance
Patch cutter	Fabricated
pH tester	Eutech instruments
Disintegration tester	Campbell electronics, Mumbai
Double beam balance	K. Roy Instruments, Hyderabad

PREFORMULATION STUDIES

Preformulation studies are vital for any kind of formulation since they assure the success of the final product both physically and chemically. The important preformulation studies with respect to this work involves:

1. Authentication of source of the Palmyra palm fruit resin
2. Preparation of the *Borassus flabellifer* fruit resin
3. Compatibility studies using FT-IR
4. Preparation of calibration graph of Metadoxine using UV-visible spectrophotometry

1. Authentication of source of the Palm fruit resin:

Various parts of the Palmyra palm such as fruits (unripened and ripened), leaf with stalk and flower were submitted for identification and authentication of the botanical source to the Botanical Survey of India, Southern Regional Centre, Coimbatore.

2. Preparation of *B. flabellifer* Fruit Resin (BFR) [25]:

A ripened fruit of *B. flabellifer* was obtained from a local vendor. The black coloured peel of the fruit was removed and the three seeds along with the fibrous pulp was partitioned. Each portion of the fruit was boiled in hot water at 40°C. The sticky, yellow pulp was manually extracted from the fibers with the help of hot water. The process was continued till the fibers were free of yellow pulp and turn into pale colour.

The seed and fibers were removed by means of filtration using a muslin cloth. The filtrate (fruit pulp) was concentrated by evaporating the liquid (at not more than 45°C), till the extract dried into a golden brown coloured sticky resin. The process of drying must be done carefully, since increase in temperature may char the product. The dried resin was stored in an air-tight container at room temperature.

3. Compatibility studies using FT-IR [35]

Compatibility studies are essential to study the interaction of the excipients with the drug, because it is an important criterion for any excipient, not to exhibit any kind of interaction with the drug. Therefore, in the present work, a study was carried out using infrared spectrophotometer to find out if there are any possible chemical interactions between drug and all the polymers used such as the new mucoadhesive polymer *B. flabellifer* fruit Resin (BFR), Pectin, Sodium alginate (SA) and PVA.

4. Preparation of calibration graph of Metadoxine using UV-visible spectrophotometry [15]

10mg of Metadoxine was dissolved in PBS pH-6.8 and the volume was made up to 100ml with the same, which gives a stock solution of 100µg/ml. From this stock solution aliquots of 0.4 – 4 ml were withdrawn using a pipette and transferred to a series of ten 10ml standard flasks. The volumes were made up with PBS pH-6.8. Thus, the concentration range of 4–40 µg/ml was obtained. The absorbances of the solutions were estimated at 324 nm using PBS pH-6.8 as reagent blank, with the help of UV-visible spectrophotometer. A triplicate of measurements was made to get mean absorbance values. A calibration graph of absorbance vs. concentration was plotted.

FORMULATION OF METADOXINE BUCCAL PATCHES

1. Optimization of polymer ratios:

Almost 50 combinations of BFR with polymers such as Carbopol-940, HPMC. HEC, PVP, Gelatin, Pectin, Sodium alginate, PVA 6000, PVA 4000, PVA 125000, PVA 160000 were tried to formulate buccal patches of formidable physical properties, by adding varying volume of plasticizer (PEG-400) and permeation enhancer (DMSO). Finally, 9 polymer ratios using Pectin, Sodium alginate and PVA-160000 were found to be suitable.

2. Dose calculation:

The average thickness of patches made up by 10ml of formulation mixture without drug, found out after a number of trials (during optimization of polymer ratios) is 0.07 cm, using a digital screw gauge. Therefore,

Volume of a parent patch made up by 10ml of formulation mixture $= 3.1429 \times 4.4 \times 4.4 \times 0.07$
$= 4.2593\ cm^3$

Volume of a single patch of radius 1cm $= 3.1429 \times 1 \times 1 \times 0.07$
$= 0.22\ cm^3$

The number of possible patches (theoretically) $= \frac{4.2593}{0.22}$
$= 19.3605$

Thus, the quantity of drug to be added $= 19.3605 \times 250\ mg$
$= 4.8401\ g$

The parent patches of each formulation were cut into uniform pieces of buccal patches of fixed diameter, using a fabricated stainless steel punch with sharp edges.

3. Formulation of buccal patches by Solvent casting method:

Weighed quantity of BFR was added to distilled water and dissolved using a magnetic stirrer set at 500 rpm to obtain a uniform solution. 12 formulations using Pectin (F1-F3), SA (F4-F6) and PVA (F7-F9) in varying proportions were added to each formulation.

The rest of the ingredients such as sucrose (sweetening agent), Vanillin (flavoring agent), PEG-400 (plasticizer) and Dimethyl sulphoxide (permeation enhancer) were added in the order as given in the Table-1. Finally, the required quantity of Metadoxine was added to the polymer matrices. The formulation mixtures were poured to petri dishes of known diameter and allowed to air-dry at room temperature, by covering the dishes with a clean sieve or in a hot air oven at 30±5 °C, till the patches form a smooth non-sticky surface.

Table-3: Composition of Metadoxine buccal patches

Formulation Code	F1	F2	F3	F4	F5	F6	F7	F8	F9
Ingredients	(in mg)								
Metadoxine	4840	4840	4840	4840	4840	4840	4840	4840	4840
BFR	300	400	500	400	400	400	300	400	500
Pectin	500	400	300	-	-	-	-	-	-
SA	-	-	-	200	300	400	-	-	-
PVA	-	-	-	-	-	-	500	400	300
Vanillin	60	60	60	60	60	60	60	60	60
Sucrose	300	300	300	300	300	300	300	300	300
	in ml								
Water	10	10	10	10	10	10	10	10	10
PEG	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
DMSO	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0

4. Application of backing membrane

A suitable backing membrane prevents the buccal patch from releasing the drug through the non-adhering side. Hence, a backing membrane consisting of 4% PVA solution was sprayed over the dried patches only on one side.

EVALUATION OF THE BUCCAL PATCHES

I. Thickness [8, 12, 36]:

Thickness must be measure before application of backing membrane. A sample patch from each formulation code was taken and measured for thickness at 5 different points using an electronic micrometer (digital screw gauge). Mean thickness and standard deviation values were calculated from the observed readings.

II. Weight variation test [8, 20, 21]:

The same condition as above, measurement before application of backing membrane is followed. A random sample of 5 patches were taken from each formulation code and their individual weights were recorded. Mean weight and standard deviation values for each formulation was calculated.

III. Folding endurance [20, 21, 23]:

Folding endurance was determined by repeatedly folding a patch at the same point till the patch breaks into halves completely. The number of times the patch was folded till the point of break is considered as a patch's folding endurance.

IV. Swelling index [36]:

Swelling index is directly related to the bioadhesive strength of a patch. One patch from each formulation code was taken in a pre-weighed basket made up of stainless steel mesh. The weights of each basket with patches were recorded. The baskets were placed in beakers; marked F1-F9; containing 4ml of PBS pH-6.8 each.

After 10mins, the baskets were removed from the beakers, residual buffer solution were thoroughly strained and the weights were again noted. Swelling index for each formulation was calculated by the following equation.

$$\text{Swelling index} = \frac{\text{Weight after swelling} - \text{Initial weight}}{\text{Initial weight}}$$

V. Surface pH[23]:

A patch from each formulation code were placed in petri dishes and they were wetted with 1ml of demineralized water, and allowed to equilibrate for 30mins. The surface pH of each patch was measured by placing the tip of the pH meter electrode on the surface of the patch and holding for at least 10mins, till the pH value attains equilibrium. The procedure was repeated twice more to obtain average surface pH and standard deviation values.

VI. Drug content assay [8, 23]:

Drug content assay was carried out by dissolving the patch completely in 50ml of PBS pH-6.8, with the help of sonicator. Then, the volumes were made up to 100ml with PBS pH-6.8. The solution is filtered. 1ml of this filtrate was further diluted to 100ml with PBS pH-6.8 and the absorbance was measured at λ_{max} of 324nm. The concentration of the solution was determined from the calibration graph, by interpolation. The drug content is determined by the following steps:

$$\text{Amount of drug present in a single patch (in mg)} = \frac{\text{Concentration from the graph} \times \text{Dilution factor}}{1000}$$

$$\text{Assay/Percentage purity} = \frac{\text{Amount of drug present}}{\text{Labelled claim}} \times 100$$

where, the dilution factor = 10000
labelled claim = 250mg

Therefore, the steps can be simplified into one equation as follows:

$$\text{Drug content \%} = \text{Concentration from graph} \times 4$$

VII. Ex-vivo bioadhesion study [36]:

a. Fabrication of the test assembly

The working double beam balance formed the basis of the fabricated bioadhesion test apparatus. The left side pan was removed and replaced with a stainless steel wire (A) of gauge 1.2mm, hung with a Teflon coated glass tube (B) of diameter 1cm, loaded with weights to equate the right side pan. The height of the total setup was adjusted to accommodate a Teflon block (E), of height 1.5cm and diameter 3.8cm with an upward protrusion of 1cm height 1.5cm diameter on one of its face, leaving a headspace of 0.5cm. The two sides were balanced so that the right side was 5g heavier than the left.

b. Measurement of adhesion force

The pig's buccal mucosa (D) was excised, washed and was tightly tied over the protrusion of the Teflon block, with the mucosal side facing upwards. The setup was placed inside a glass beaker(F) with sufficient quantity of PBS pH-6.8, such that the buffer reaches the surface of the mucosal membrane and keeps it moist. This beaker was placed inside the left side of the balance. A patch (C) was stuck onto the Teflon coated tube (B) with a drop of water and the beam is raised by removing the 5g weight from the right side pan. This lowered the Teflon coated tube (B) along with the patch over the mucosa, with a weight of 5g. The balance was kept in this position for 3mins and then weights were added gradually on the right pan till the patch gets separated from the mucosal surface completely. The excess weights of the pan i.e., the total weight subtracted by 5, gives the measure of force of detachment of the patch in grams. From this the bioadhesion strength can be calculated by

$$\text{Force of adhesion (N)} = \frac{\text{Force of detachment}}{1000} \times 9.81$$

The procedure was repeated for one patch from each formulation code. A fresh portion of tissue was used for each measurement.

Fig-6: Fabricated bioadhesion test assembly

A – Stainless steel wire

B – Teflon coated glass tube with weights

C – Metadoxine buccal patch

D – Pig buccal mucosa tissue

E – Teflon block

F – Glass beaker

The results of all the above evaluation tests are given in Table-7.

VIII. In-vitro diffusion/permeation study [23]:

In-vitro drug diffusion studies were performed by using Franz diffusion cell. It consists of a donor compartment and a receptor compartment. The receptor compartment is filled with 16ml of PBS pH-6.8 as the diffusion medium along with a magnetic bead. Over the filled receptor compartment, cellulose nitrate membrane of pore size 0.2µm was placed and allowed to moisten for 1min, to mimic buccal mucosa environment. Then a patch under study was placed over the membrane and closed tightly with the donor compartment.

The whole assembly is fixed over a hot plate magnetic stirrer and the medium in the receptor compartment was subjected to stirring a 100rpm and the temperature of the diffusion cell is supplied constantly with flowing hot water at 37°±1° C to simulate the fluid and thermodynamics of the buccal environment.

Fig-7: Franz Diffusion cell

One ml samples were withdrawn from the sample port at predetermined time intervals with the help of a 1ml disposable syringe and the same volume was replaced with PBS pH-6.8. The samples were suitably diluted with the same medium and are analyzed for drug content at 324 nm, using PBS pH 6.8 as reagent blank. The unknown concentrations of the samples were obtained from the calibration graph of Metadoxine. The procedure is repeated for a sample patch from all formulations.

The cumulative percentage release values for the respective time are tabulated (Table-8) and cumulative percentage release (%) vs time plots are drawn (Figures 20-22).

IX. In-vitro drug release kinetics [37]

The order and mechanism of drug release kinetics of Metadoxine buccal patches were analyzed using the in-vitro diffusion study data, by plotting different kinetic models such as zero order, first order and Higuchi equations. The release pattern was determined using Korsmeyer-Peppas equations.

a. Zero order kinetics model

Cumulative percentage of drug diffused was plotted against time.

$$\mathbf{Q=K_0t}$$

where $\mathbf{K_0}$ is the zero order rate constant expressed in unit percentage of drug diffused (**Q**) /time (**t**) in hours. A graph of cumulative % drug diffused vs. time would yield a straight line with a slope K_0 and intercept the origin of the axis. This kinetic model describe that the drug diffusion is concentration independent.

b. First order kinetics model

The pharmaceutical formulations following this kinetic model, release the drug in a way that is proportional to the amount of drug remaining in its interior, in such a way, that the amount of drug released diminish with time. First order kinetics graph is obtained by plotting log cumulative % drug diffused vs time. This kinetic model describe that the diffusion is concentration dependent.

$$\mathbf{\log Q = \log Q_0 - K_1t / 2.303}$$

where **Q** is the cumulative % drug diffused at time '**t**'

$\mathbf{Q_0}$ is the cumulative % drug diffused at '0' time

$\mathbf{K_1}$ is the rate constant of first order kinetics

c. Higuchi's model

Higuchi's model is based on a plot of cumulative percentage of drug released vs. square root of time.

$$\mathbf{Q = K_H.t^{1/2}}$$

where $\mathbf{K_H}$ is the constant reflecting the design variables of the system and '**t**' is the time in hours. This model describes the release of drug on the basis of Fickian diffusion as a square root of time dependent process from swellable matrix.

d. Korsmeyer-Peppas Equations

Korsmeyer-Peppas equation is used to evaluate the release pattern, by using the equation

$$\mathbf{Q = K_{kp} t^n}$$

where **Q** is the fractional solute release, **t** is the release time, $\mathbf{K_{kp}}$is a kinetic constant characteristic of the drug/polymer system, and **n** is an exponent that characterizes the mechanism of release.

The equation is subjected to modification by taking log on both sides, thereby the equation is

$$\mathbf{\log Q = \log K_{kp} + n \log t}$$

The exponent '**n**' can be calculated through the slope of the linear graph of log cumulative percentage of drug released (log Q) vs. log time (log t). The 'n' value is used to characterize the diffusion mechanism based on the data given in Table-4.

Table-4: Diffusion exponent and diffusion mechanism

Diffusion exponent	Overall diffusion mechanism
0.5	Quasi Fickian diffusion
0.5	Fickian diffusion (Higuchi Matrix)
0.5 n < 1.0	Non-Fickian diffusion
1.0	Case 2 transport
>1.0	Super case 2 transport

Softwares such as DD Solver and Kinet DS are specifically programmed for calculating kinetic models. In this study, DD Solver was used to propagate respective graphs (Figures:23-31) of each model, using cumulative percentage release per time data.

RESULTS & DISCUSSIONS

I. PREFORMULATION

1. Authentication of source of the Palmyra palm fruit resin

The source of the Palmyra palm resin was authenticated as the fruit pulp of *Borassus flabellifer*. L, belonging to family Arecaceae.

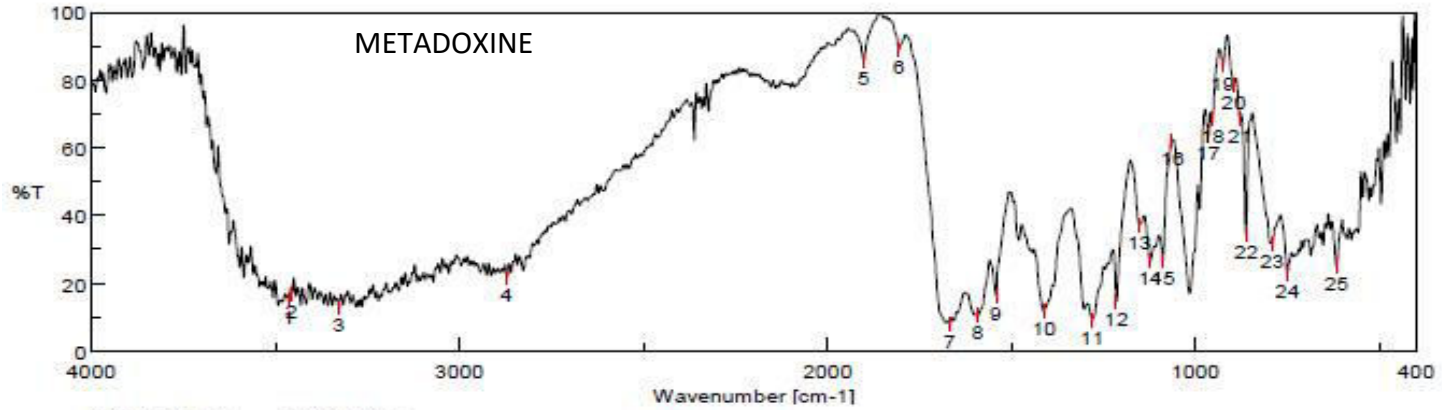
2. Preparation of the *Borassus flabellifer* fruit resin

Fig-8: *B. flabellifer* Fruit Resin

3. Compatibility studies using FT-IR [30]

The physical mixtures of Metadoxine and polymers were subjected to FT-IR analysis to identify any interaction between them.

FT-IR spectra of Metadoxine, BFR, Pectin, Sodium alginate, PVA and mixtures of drug with each excipient are given in Figures 9-7.

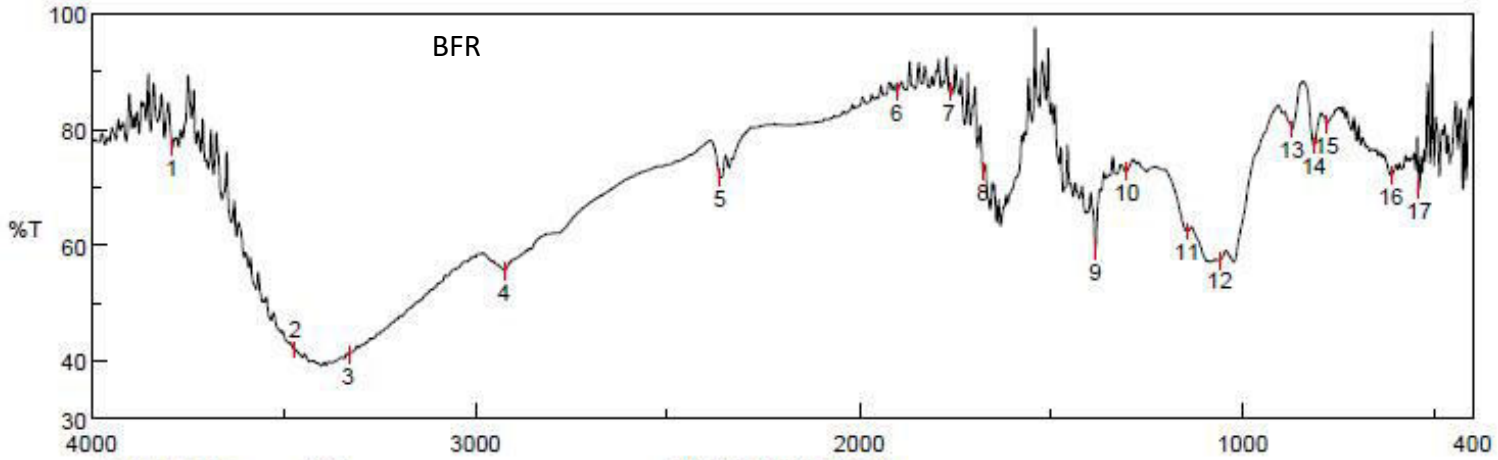


Sample name	Metadoxine
Comment	FTIR SPECTRUM
User	V. Hariharshan
Division	Pharmaceutics
Company	SRIPMS

[Result of Peak Picking]

No.	Position	Intensity	No.	Position	Intensity	No.	Position	Intensity
1	3462.56	14.8127	2	3454.85	16.9458	3	3327.57	12.6382
4	2870.52	21.8789	5	1900.5	85.8196	6	1806.01	89.1146
7	1667.16	7.93445	8	1590.02	10.4831	9	1541.81	16.2447
10	1412.6	11.6029	11	1281.47	9.22126	12	1214.93	14.5265
13	1152.26	37.252	14	1123.33	26.6095	15	1088.62	26.7002
16	1062.59	61.8441	17	965.198	63.5685	18	953.627	68.4923
19	925.664	84.3324	20	895.773	78.3626	21	879.381	68.8516
22	862.989	34.4478	23	791.636	31.2943	24	750.174	22.6344
25	617.109	25.1174						

Fig-9: Infrared spectrum of Metadoxine

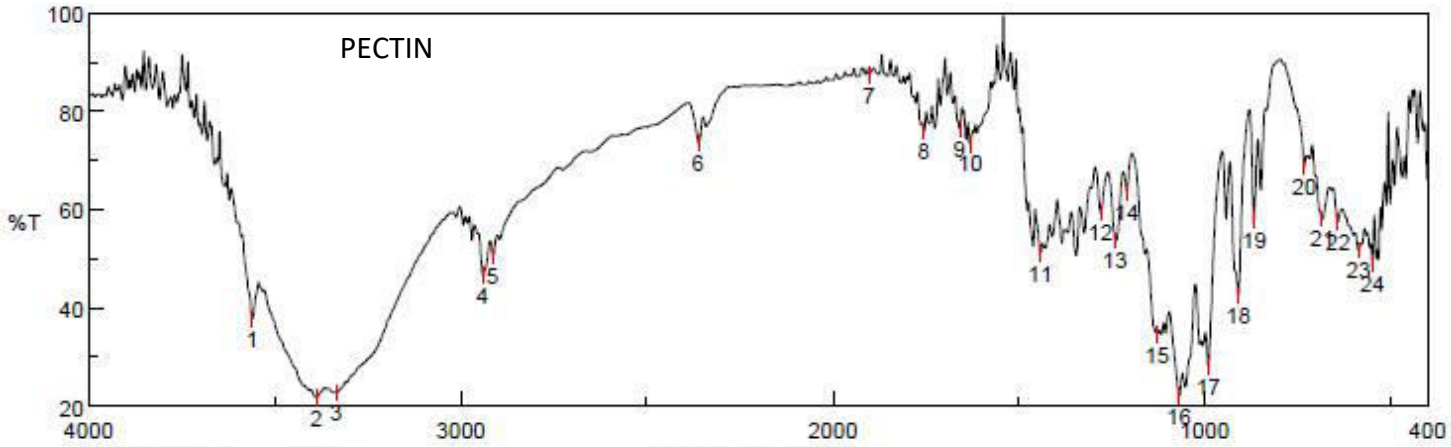


Sample name	BFR
Comment	FTIR SPECTRUM
User	V. Hariharshan
Division	Pharmaceutics
Company	SRIPMS

[Result of Peak Picking]

No.	Position	Intensity	No.	Position	Intensity	No.	Position	Intensity
1	3790.4	76.8053	2	3470.28	41.9556	3	3331.43	40.9884
4	2924.52	55.5269	5	2362.37	71.6328	6	1900.5	86.4858
7	1764.55	86.4219	8	1675.84	72.7281	9	1383.68	59.0988
10	1302.68	72.7728	11	1145.51	62.3764	12	1059.69	57.2991
13	870.703	79.931	14	813.813	77.0755	15	781.029	80.8795
16	613.252	71.9293	17	539.971	69.659			

Fig-10: Infrared spectrum of BFR

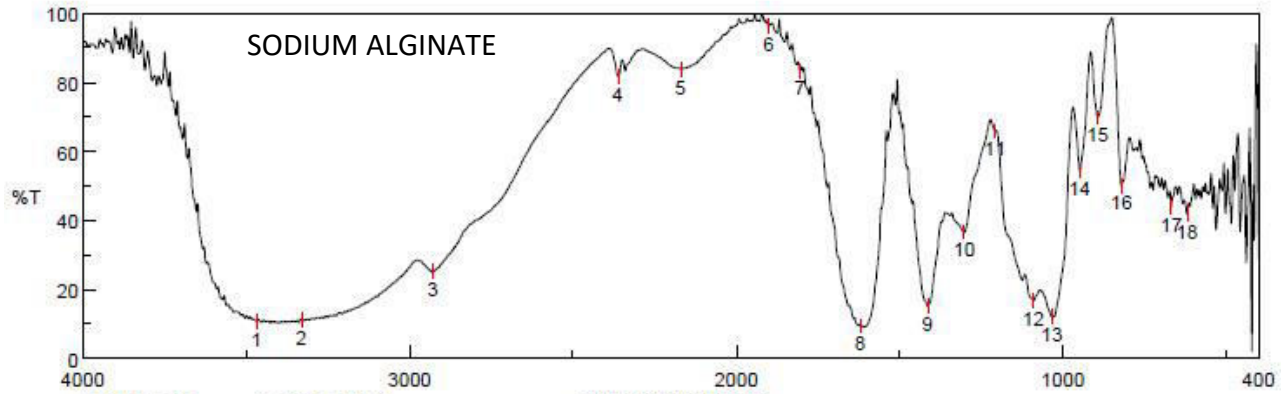


Sample name	Pectin
Comment	FTIR SPECTRUM
User	V. Hariharshan
Division	Pharmaceutics
Company	SRIPMS

[Result of Peak Picking]

No.	Position	Intensity	No.	Position	Intensity	No.	Position	Intensity
1	3560.91	37.7819	2	3387.35	21.7705	3	3335.28	22.7849
4	2940.91	46.5858	5	2912.95	50.5051	6	2361.41	73.5793
7	1900.5	87.4753	8	1754.9	75.7962	9	1658.48	76.2426
10	1630.52	73.3814	11	1441.53	51.1434	12	1279.54	59.3363
13	1240	53.6831	14	1209.15	63.4013	15	1128.15	34.6971
16	1068.37	22.0105	17	990.268	28.0671	18	909.272	42.544
19	866.846	57.8968	20	731.853	68.7684	21	683.642	58.0703
22	641.215	57.5301	23	586.254	51.6666	24	548.649	49.1845

Fig-11: Infrared spectrum of Pectin

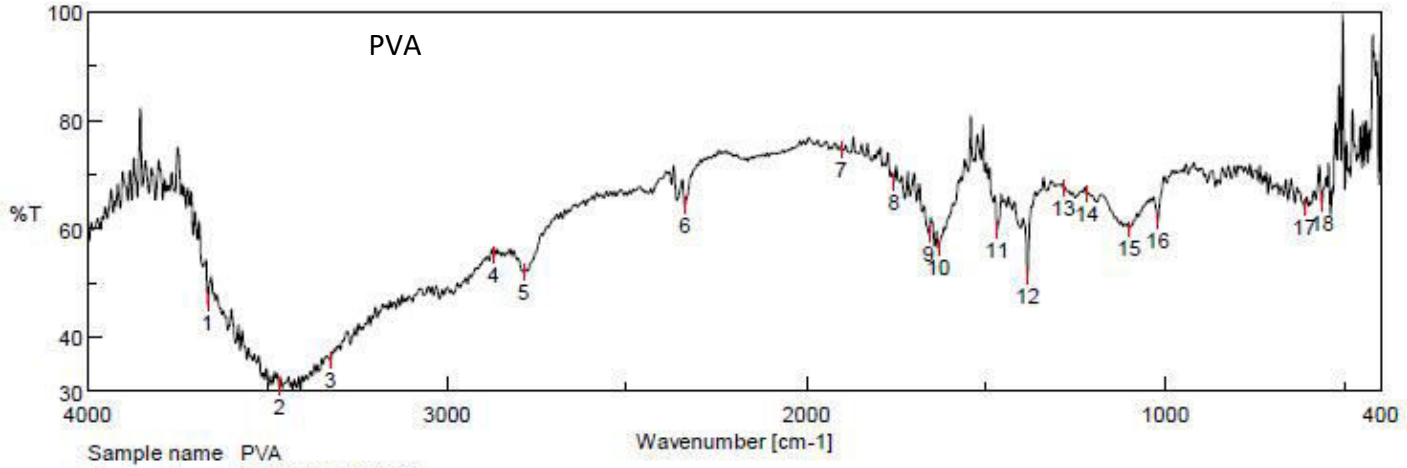


Sample name	Sodium alginate
Comment	FTIR SPECTRUM
User	V. Hariharshan
Division	Pharmaceutics
Company	SRIPMS

[Result of Peak Picking]

No.	Position	Intensity	No.	Position	Intensity	No.	Position	Intensity
1	3468.35	10.7619	2	3330.46	11.1209	3	2927.41	25.1818
4	2360.44	81.7831	5	2169.53	83.8308	6	1899.54	96.3163
7	1805.05	83.3923	8	1619.91	9.43778	9	1413.57	15.2202
10	1302.68	36.5583	11	1207.22	65.8433	12	1089.58	16.7736
13	1034.62	12.1179	14	947.842	54.256	15	892.88	69.9241
16	820.563	50.1878	17	668.214	44.0884	18	620.002	41.8706

Fig-12: Infrared spectrum of Sodium alginate

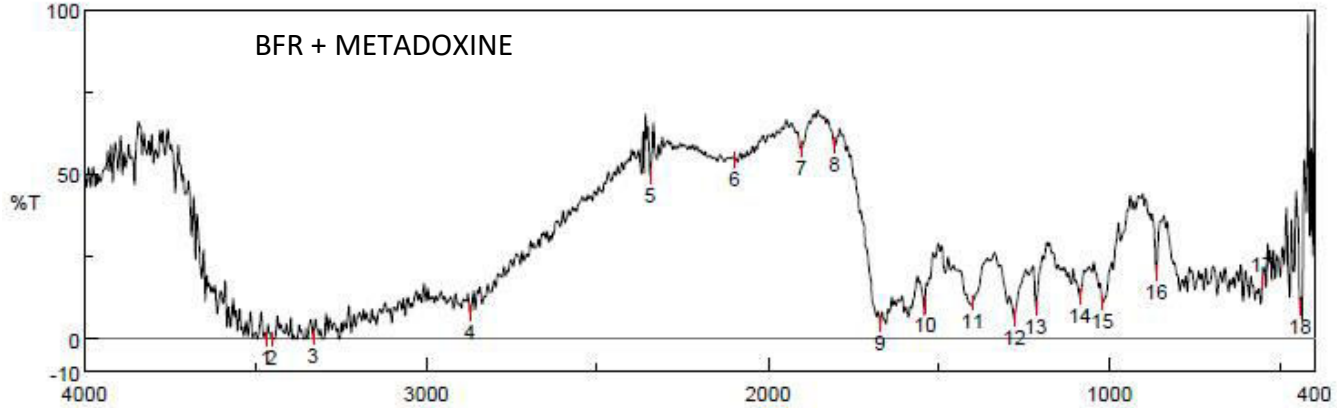


Sample name	PVA
Comment	FTIR SPECTRUM
User	V. Hariharshan
Division	Pharmaceutics
Company	SRIPMS

[Result of Peak Picking]

No.	Position	Intensity	No.	Position	Intensity	No.	Position	Intensity
1	3666.02	46.1829	2	3467.38	30.8556	3	3324.68	35.6473
4	2869.56	55.0174	5	2785.67	51.8884	6	2338.27	64.2652
7	1902.43	74.3568	8	1754.9	68.5877	9	1658.48	58.9758
10	1631.48	56.3449	11	1468.53	59.8002	12	1383.68	51.0178
13	1284.36	67.4935	14	1218.79	66.3433	15	1101.15	59.9071
16	1021.12	61.455	17	612.288	63.8977	18	564.077	64.5215

Fig-13: Infrared spectrum of PVA

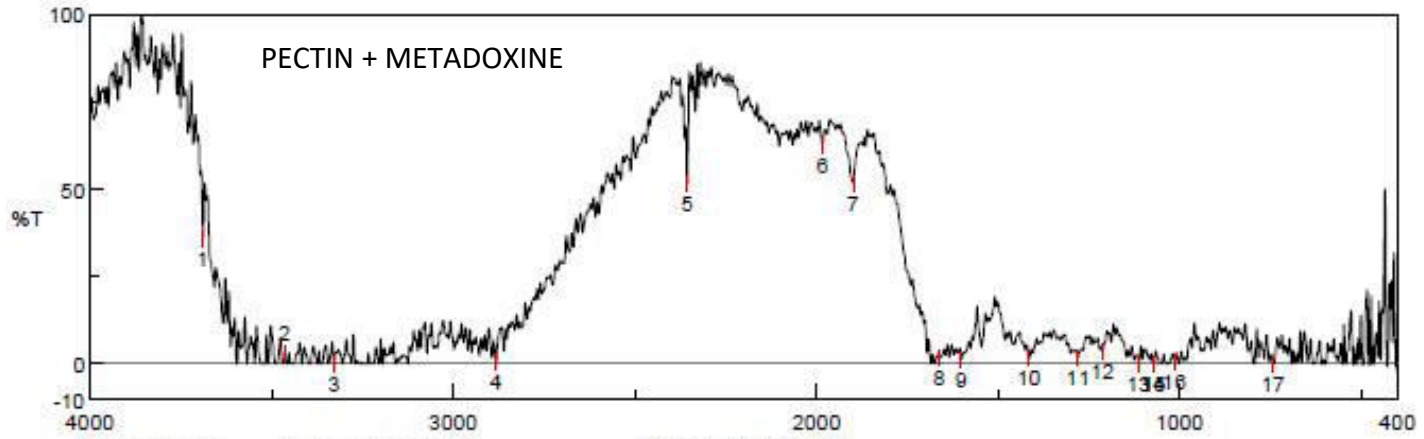


Sample name	BFR + Metadoxine
Comment	FTIR SPECTRUM
User	V. Hariharshan
Division	Pharmaceutics
Company	SRIPMS

[Result of Peak Picking]

No.	Position	Intensity	No.	Position	Intensity	No.	Position	Intensity
1	3468.35	-0.158232	2	3452.92	-0.0431322	3	3330.46	0.328152
4	2871.49	7.74983	5	2344.05	49.4861	6	2098.17	54.2276
7	1901.47	57.6053	8	1804.08	58.7395	9	1671.02	4.69389
10	1539.88	10.0426	11	1401.03	10.9443	12	1278.57	6.11605
13	1214.93	9.15626	14	1086.69	12.9692	15	1020.16	10.8578
16	862.989	19.9997	17	553.47	17.0394	18	442.583	9.60199

Fig-14: Infrared spectrum of BFR + Metadoxine

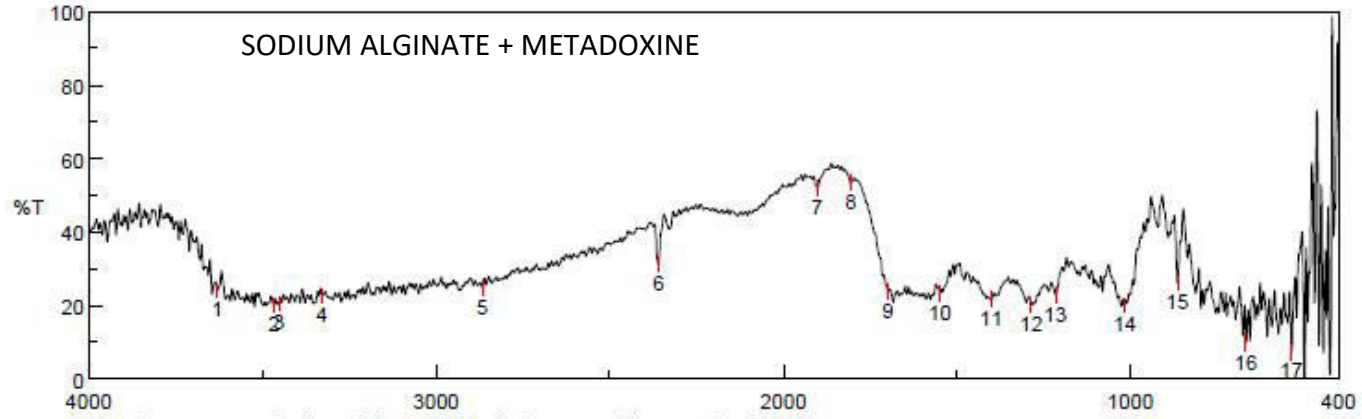


Sample name	Pectin + Metadoxine
Comment	FTIR SPECTRUM
User	V. Hariharshan
Division	Pharmaceutics
Company	SRIPMS

[Result of Peak Picking]

No.	Position	Intensity	No.	Position	Intensity	No.	Position	Intensity
1	3689.16	35.3418	2	3463.53	3.01144	3	3328.53	-0.150572
4	2881.13	0.321803	5	2355.62	51.4761	6	1983.43	62.5594
7	1898.58	51.6545	8	1661.37	1.45341	9	1600.63	1.22201
10	1414.53	1.52899	11	1280.5	1.71187	12	1212.04	3.52939
13	1112.73	0.145441	14	1073.19	-0.113996	15	1070.3	-0.106364
16	1014.37	0.602135	17	742.46	-0.0842225			

Fig-15: Infrared spectrum of Pectin+ Metadoxine

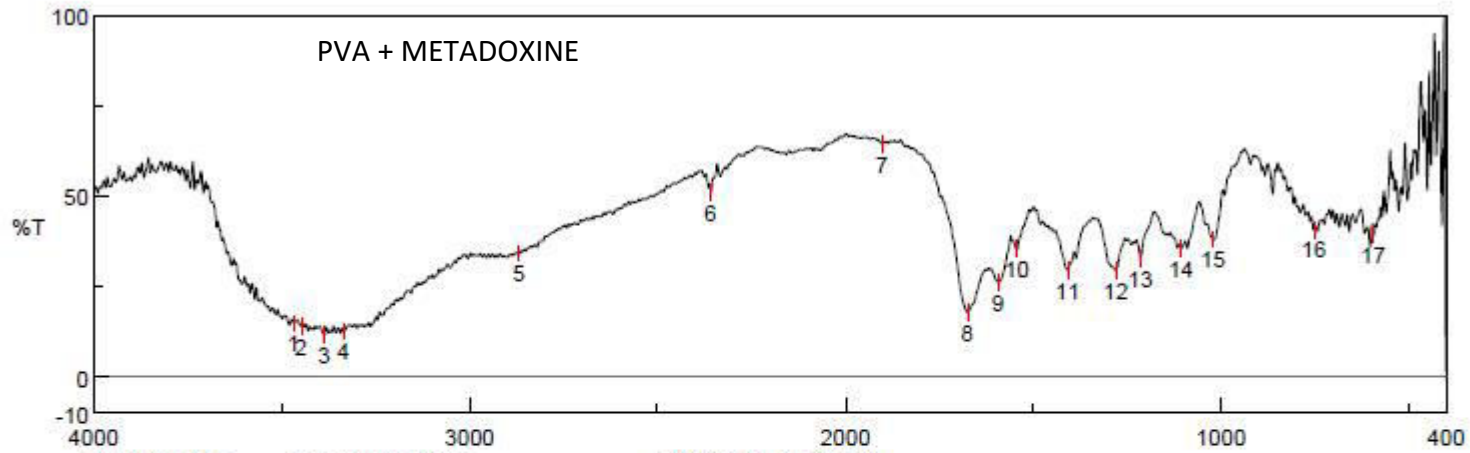


Sample name Sodium alginate + Metadoxine Wavenumber [cm-1]
Comment FTIR SPECTRUM
User V. Hariharshan
Division Pharmaceutics
Company SRIPMS

[Result of Peak Picking]

No.	Position	Intensity	No.	Position	Intensity	No.	Position	Intensity
1	3629.37	24.126	2	3467.38	20.0808	3	3452.92	20.5814
4	3327.57	22.5372	5	2866.67	24.7324	6	2358.52	31.0336
7	1900.5	52.0218	8	1804.08	53.4838	9	1697.05	23.8046
10	1548.56	23.1921	11	1401.03	21.616	12	1285.32	20.0238
13	1214.93	22.8806	14	1016.3	19.9382	15	863.953	26.083
16	667.25	9.82255	17	535.15	7.38935			

Fig-16: Infrared spectrum of Sodium alginate + Metadoxine



Sample name	PVA + Metadoxine
Comment	FTIR SPECTRUM
User	V. Hariharshan
Division	Pharmaceutics
Company	SRIPMS

[Result of Peak Picking]

No.	Position	Intensity	No.	Position	Intensity	No.	Position	Intensity
1	3468.35	14.9289	2	3448.1	14.0884	3	3386.39	11.6452
4	3335.28	12.498	5	2867.63	34.1578	6	2357.55	51.0903
7	1901.47	64.439	8	1673.91	17.7766	9	1592.91	26.3228
10	1541.81	35.4085	11	1407.78	29.6707	12	1279.54	29.5792
13	1213.97	32.7183	14	1105.98	35.683	15	1021.12	37.9341
16	753.066	40.6833	17	593.968	39.1576			

Fig-17: Infrared spectrum of PVA + Metadoxine

Table-5: Interpretation of IR spectra of drug, polymers & physical mixtures

Functional group assignment	Standard wave number (cm^{-1})	Test wave number of Metadoxine (cm^{-1})	Test wave number of polymers (cm^{-1})				Test wave number of mixtures (cm^{-1})			
			BFR	Pectin	Sodium alginate	PVA	BFR + Drug	Pectin + Drug	Sodium alginate + Drug	PVA + Drug
O-H stretching	3200-3550	3462.56	3470.28	3531.99	3468.35	3467.38	3468.35	3463.53	3467.38	3468.35
N-H stretching (aliphatic)	3310-3350	3327.57	-	-	-	-	3330.46	3328.53	3327.57	3335.28
C=O stretching	2500-3300	2870.52	2924.52	2912.95	2927.41	2869.56	2871.49	2881.13	2866.67	2867.63
C-H bending	1650-2000	1900.5	1900.5	1900.5	1899.54	1902.43	1901.47	1898.58	1900.5	1901.47
C=O stretching	1705-1725	1667.16	1675.84	1658.48	1656.55	1658.48	1671.02	1661.37	1697.05	1673.91
N-H stretching (aromatic)	1266-1342	1281.47	-	-	-	-		1280.5	1285.32	1279.54

Wavenumbers for individual compounds and physical mixtures were compared in Table-5. There was no appearance or disappearance of any characteristic peak of the drug, which confirms the absence of chemical interaction between drug and the polymers.

4. Preparation of calibration graph of Metadoxine using UV-visible spectrophotometry

The mean absorbance values for the standard concentrations of Metadoxine are given in the table-. It was found that the concentration of Metadoxine in the range 4-40µg/ml obeyed Beer-Lambert's law. The correlation coefficient was found to be 0.997862.

Table-6: Calibration graph of Metadoxine

S. no	Concentration (µg/ml)	Absorbance
1	4	0.1587
2	8	0.1954
3	12	0.3350
4	16	0.4220
5	20	0.5418
6	24	0.6303
7	28	0.7253
8	32	0.8514
9	36	0.9826
10	40	1.0630

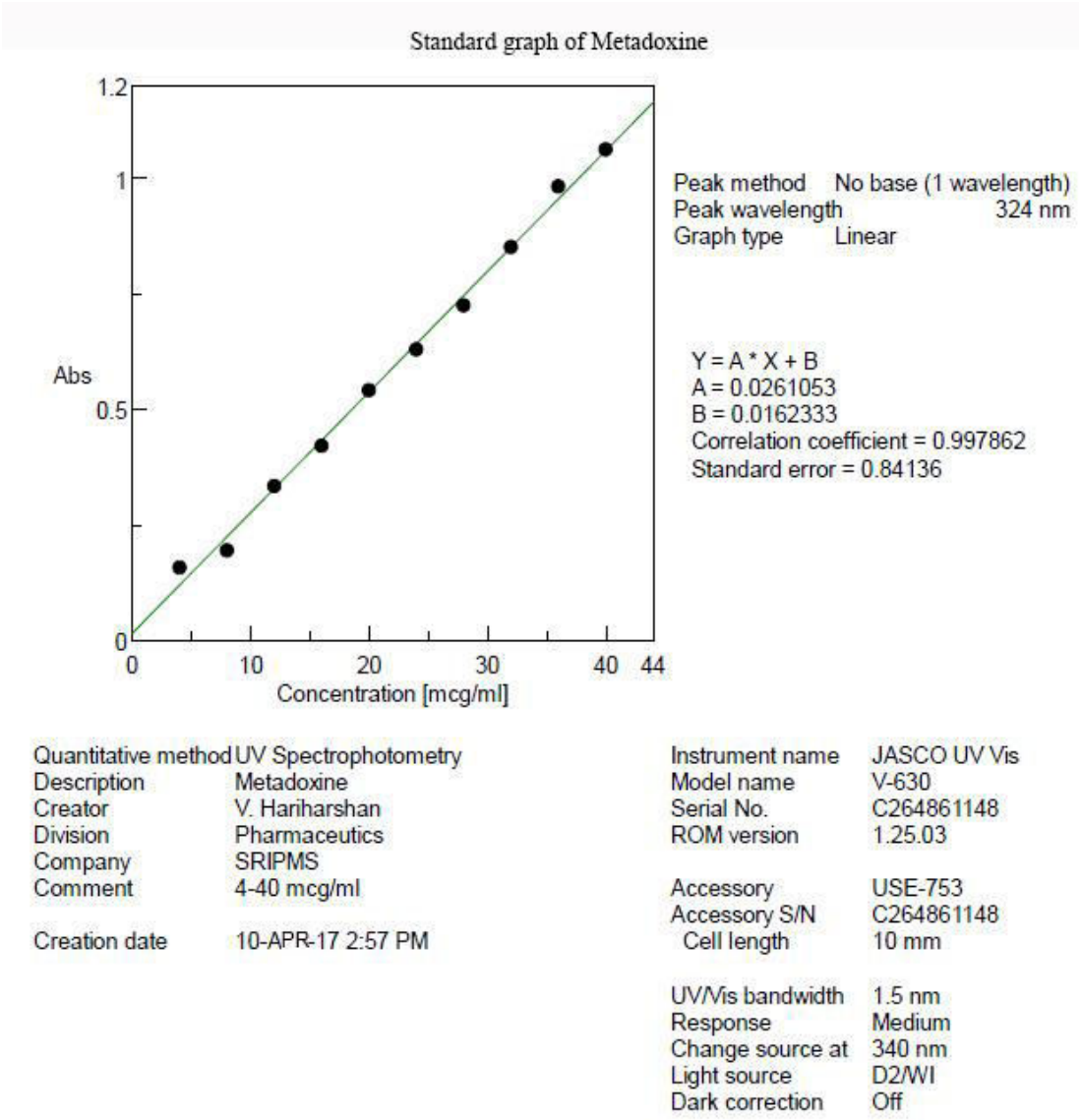
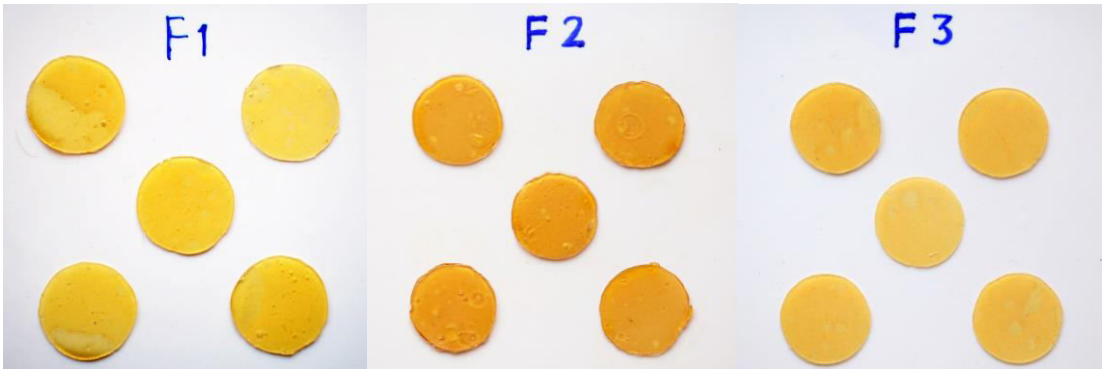


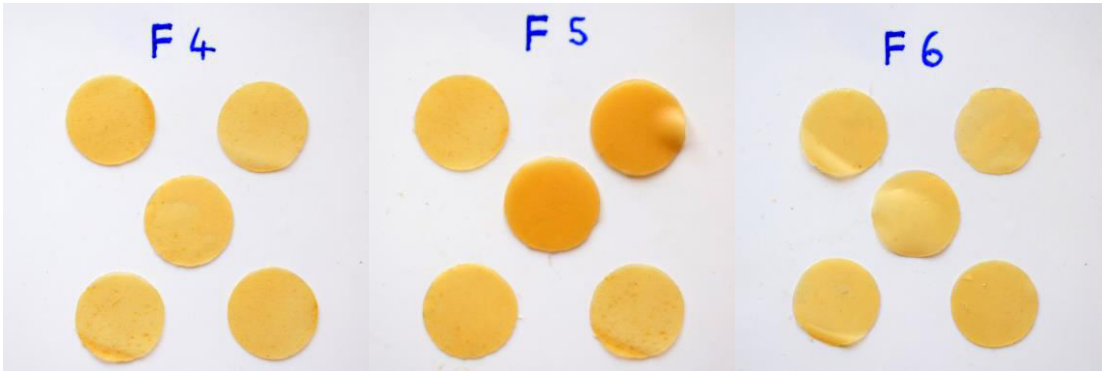
Fig-18: Calibration graph of Metadoxine

II. FORMULATION OF METADOXINE BUCCAL PATCHES

1. Formulations F1-F3: combination of BFR + Pectin



2. Formulations F4, F5 & F6: combination of BFR + Sodium alginate



3. Formulations F7, F8 & F9: combination of BFR + PVA

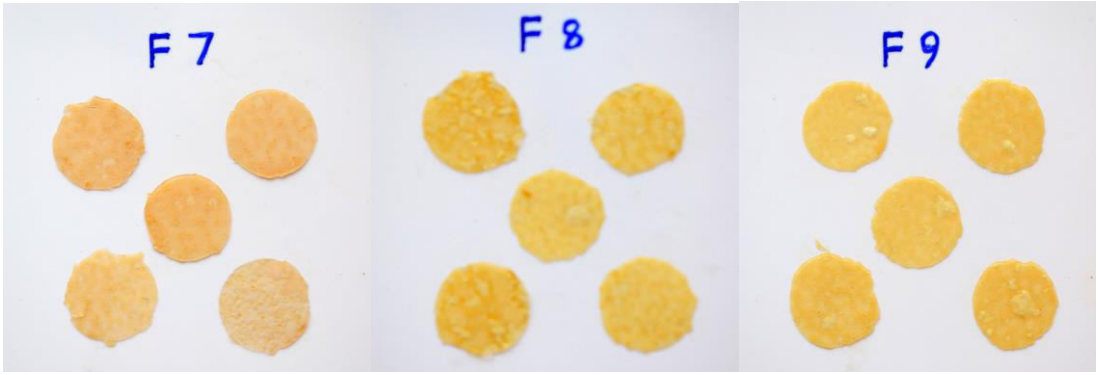


Fig-19: Photographs of Metadoxine buccal patches

III. EVALUATION OF METADOXINE BUCCAL PATCHES

A. Evaluation of physico-chemical properties

The results of physico-chemical evaluation tests such as thickness, weight variation, folding endurance swelling index, surface pH, drug content assay and bioadhesion strength are given as follows:

Table-7: Physico-chemical evaluation test results of Metadoxine buccal patches F1-F9

Formulation code	Thickness (mm)	Weight variation (mg)	Folding endurance	Swelling Index	Surface pH	Bioadhesion strength (N)	Drug content assay (%)
F1	0.7318 ± 0.02	425.8 ± 3.77	61	3.8125	6.83 ± 0.1	0.0183	97.6
F2	0.7294 ± 0.03	383.6 ± 4.39	16	0.6279	6.56 ± 0.08	0.0086	94
F3	0.6882 ± 0.02	399.6 ± 3.84	53	0.5152	6.51 ± 0.34	0.0398	90
F4	0.6978 ± 0.01	343.4 ± 4.21	56	4.0909	7.34 ± 0.09	0.0256	95.2
F5	0.7536 ± 0.01	350.2 ± 4.32	81	2.5857	6.84 ± 0.06	0.0360	96.8
F6	0.7190 ± 0.09	361.2 ± 3.11	152	4.0667	5.99 ± 0.11	0.0392	85.6
F7	0.7658 ± 0.02	399.8 ± 3.11	256	1.5455	7.17 ± 0.13	0.0187	99.6
F8	0.7152 ± 0.06	390.6 ± 3.28	230	0.6154	7.06 ± 0.09	0.0144	100.8
F9	0.6912 ± 0.03	386.8 ± 4.43	178	0.3571	6.89 ± 0.04	0.0271	96

Inference

- The thickness of the patches ranges from 0.6882 ± 0.02 mm to 0.7658 ± 0.02 mm, which ascertains that the average thickness assumed(0.7mm) for dose calculation is valid.

- The weighs of the patches of different formulation codes were in the range of 343.4 ± 4.21 mg to 425.8 ± 3.77 mg, whereas the intra-batch variation is relatively smaller with a maximum standard deviation of 4.43 mg (F9).

- The patches F7-F9 exhibited remarkable folding endurance with values as high as 256, whereby the lowest value of 16 was observed for F2. Increase in the additional polymer (Pectin/SA/PVA) increases the folding endurance,

- Swelling index of all the formulations were relatively good, with highest swelling property exhibited by F4 (BFR : SA - 4:2) at 4.099.

- The surface pH values of the formulations were in the range 5.99 ± 0.11 to 7.34 ± 0.09, which indicates the patches have a similar pH to that of saliva (pH-6.8) and thus they will not irritate the buccal mucosa. A decrease in pH was observed with increase in BFR concentration, which is due to the inherent pH (5.5) of the polymer itself.

- The force required to detach the patch from the animal's tissue is directly proportional to the bioadhesion strength of the patches. In this aspect, the patch with highest bioadhesion strength (0.0398 N) was exhibited by F3 (BFR : Pectin – 5:3). This indicates that high concentration of BFR can help to retain the patch over the mucosa for a longer period, in spite of the mechanics of the facial tissues.

- The test for drug content resulted in assay values as high as 100.8 % w/w and not less than 85.6% w/w, which proves that the method employed for formulation and dose calculation were appropriate and has good reproducibility.

B. Evaluation of percentage drug release

Table-8: In-vitro permeation data of formulations F1, F2 & F3

S.no	Time (h)	Cumulative percentage release %		
		F1	F2	F3
1	0	0	0	0
2	0.5	1.64	3.49	3.57
3	1.0	6.56	13.98	5.65
4	1.5	12.96	15.63	7.143
5	2.0	18.71	17.84	16.58
6	2.5	19.69	37.84	31.43
7	3.0	20.35	42.16	50.71
8	3.5	38.73	47.78	58.73
9	4.0	46.54	55.09	63.21
10	4.5	59.58	69.32	70.22
11	5.0	73.9	79.36	75.68
12	5.5	80.6	87.23	86.98
13	6.0	92.6	96.84	91.23

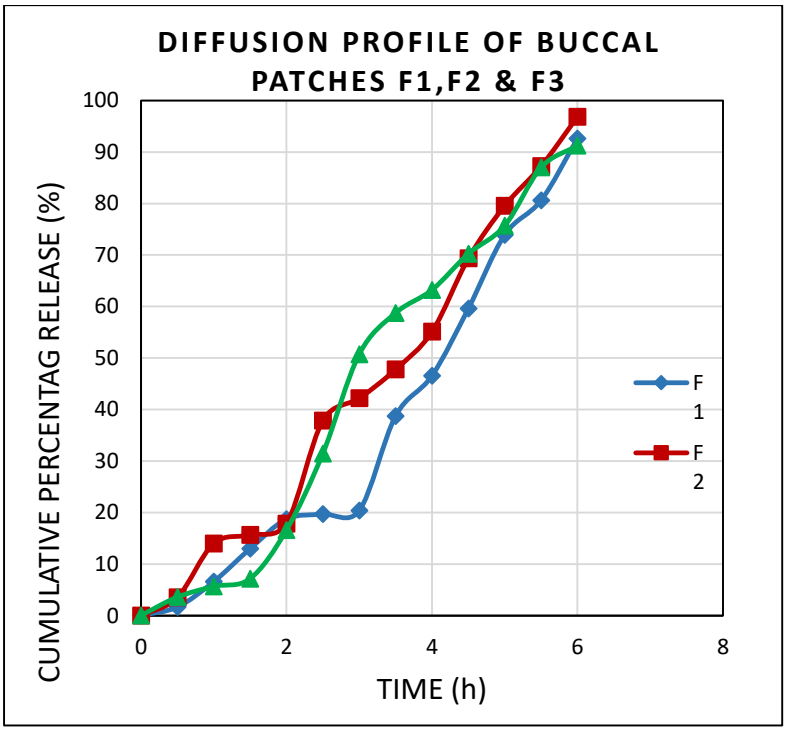


Fig-20: In-vitro diffusion profile of formulations F1, F2 & F3

Table-9: In-vitro permeation data of formulations F4, F5 & F6

S.no	Time (h)	Cumulative percentage release %		
		F4	F5	F6
1	0	0	0	0
2	0.5	1.56	2.12	2.73
3	1.0	4.24	5.57	3.07
4	1.5	15.93	18.39	4.76
5	2.0	28.75	26.98	5.24
6	2.5	37.98	42.61	9.98
7	3.0	51.69	54.00	12.4
8	3.5	65.54	63.21	24.77
9	4.0	74.65	70.02	38.68
10	4.5	83.62	81.09	52.32
11	5.0	88.82	85.64	72.26
12	5.5	-	90.08	85.41
13	6.0	-	93.21	-

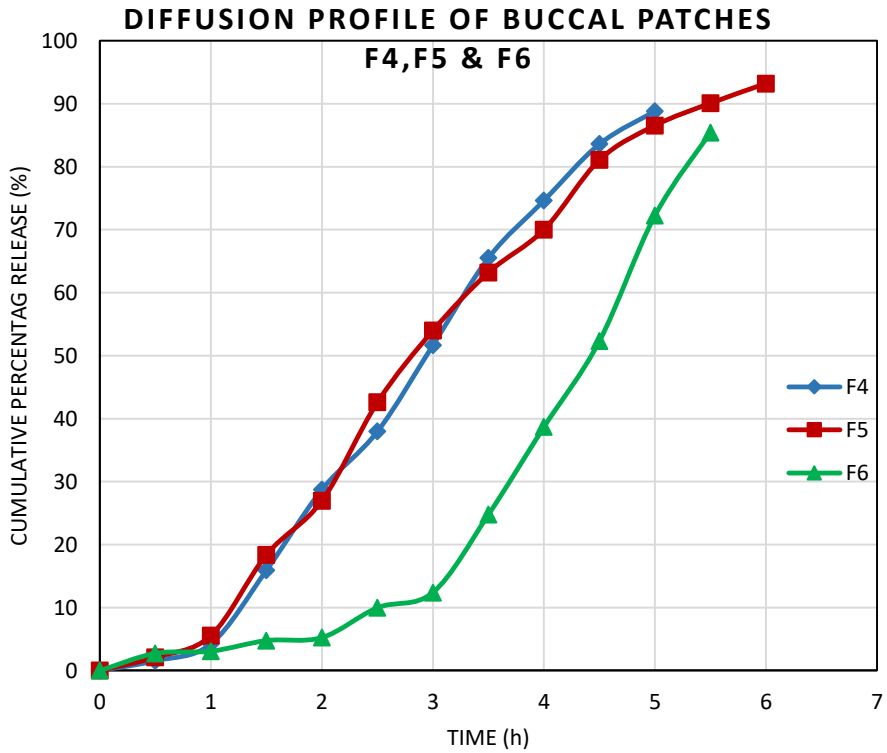


Fig-21: In-vitro diffusion profile of formulations F4, F5 & F6

Table-10: In-vitro permeation data of formulations F7, F8 & F9

S.no	Time (h)	Cumulative percentage release %		
		F7	F8	F9
1	0	0	0	0
2	0.5	1.72	4.15	6.86
3	1.0	3.26	6.23	9.18
4	1.5	10.32	11.59	11.52
5	2.0	10.81	23.56	15.08
6	2.5	12.17	39.58	31.83
7	3.0	14.04	40.21	36.19
8	3.5	33.14	47.65	40.86
9	4.0	36.61	59.10	47.98
10	4.5	40.29	76.09	58.65
11	5.0	55.09	88.64	64.72
12	5.5	63.74	95.12	73.69
13	6.0	72.35	-	88.51

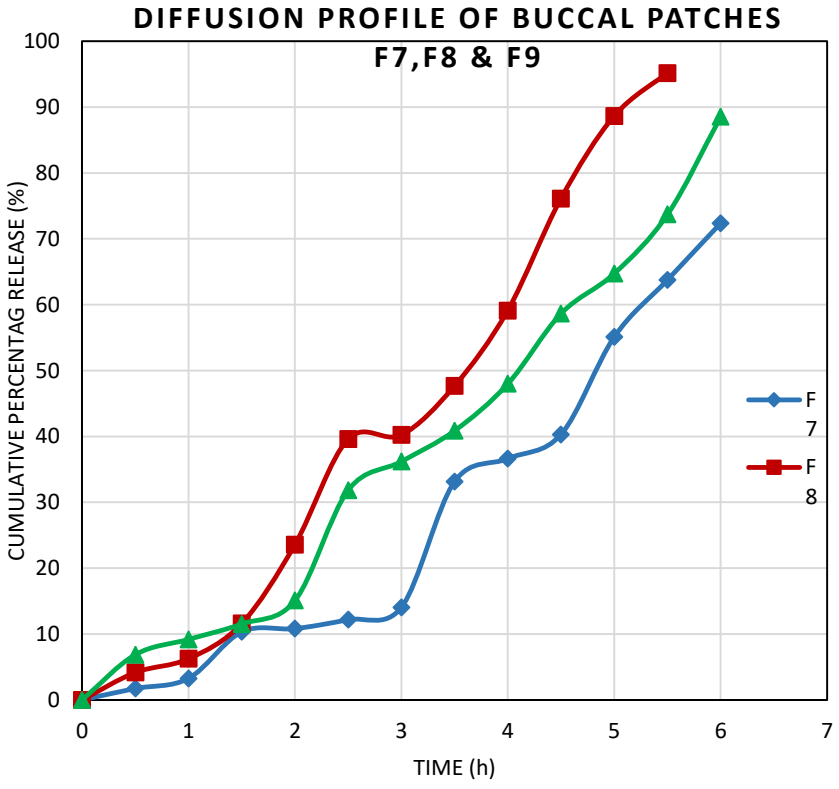


Fig-22: In-vitro diffusion profile of formulations F7, F8 & F9

Table-11: In-vitro permeation data of formulations F1- F9

S.no	Time (h)	Cumulative percentage release %								
		F1	F2	F3	F4	F5	F6	F7	F8	F9
1	0	0	0	0	0	0	0	0	0	0
2	0.5	1.64	3.49	3.57	1.56	2.12	2.73	1.72	4.15	6.86
3	1.0	6.56	13.98	5.65	4.24	5.57	3.07	3.26	6.23	9.18
4	1.5	12.96	15.63	7.143	15.93	18.39	4.76	10.32	11.59	11.52
5	2.0	18.71	17.84	16.58	28.75	26.98	5.24	10.81	23.56	15.08
6	2.5	19.69	37.84	31.43	37.98	42.61	9.98	12.17	39.58	31.83
7	3.0	20.35	42.16	50.71	51.69	54.00	12.4	14.04	40.21	36.19
8	3.5	38.73	47.78	58.73	65.54	63.21	24.77	33.14	47.65	40.86
9	4.0	46.54	55.09	63.21	74.65	70.02	38.68	36.61	59.10	47.98
10	4.5	59.58	69.32	70.22	83.62	81.09	52.32	40.29	76.09	58.65
11	5.0	73.9	79.36	75.68	88.82	85.64	72.26	55.09	88.64	64.72
12	5.5	80.6	87.23	86.98	-	90.08	85.41	63.74	95.12	73.69
13	6.0	92.6	96.84	91.23	-	93.21	-	72.35	-	88.51

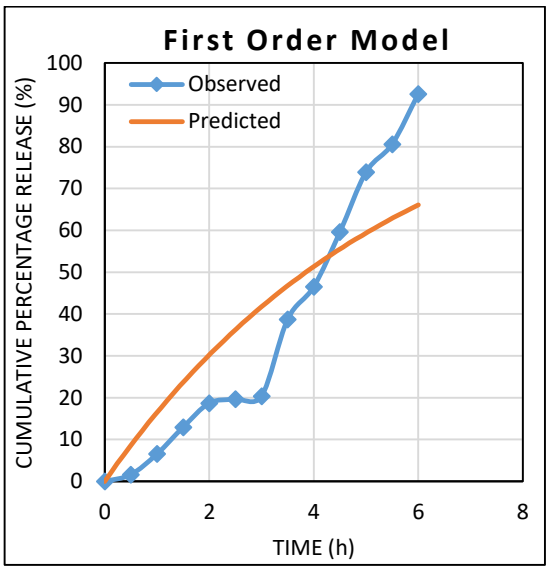
C. Release kinetics

The results of drug release kinetics study were tabulated on the basis of in-vitro diffusion study data.

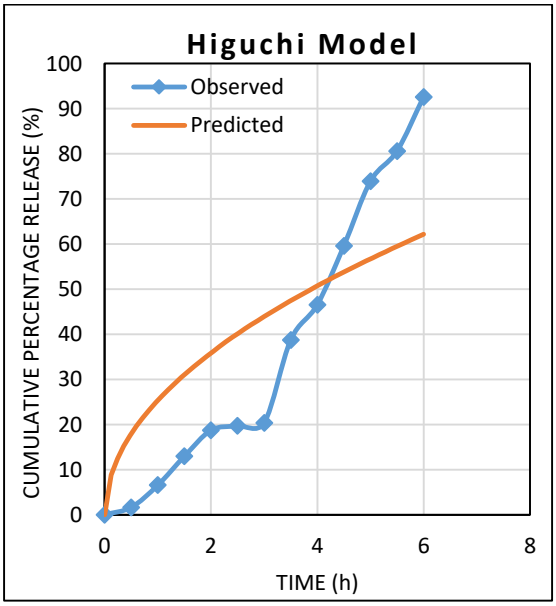
Table-12: Release kinetics data of F1 [BFR : Pectin – 3:5]

S.no	Time-t (h)	Square root of time - $t^{1/2}$	log t	Cumulative percentage of drug diffused-Q (%)	log Q
1	0	0	0	0	0
2	0.5	0.7071	-0.3010	1.64	0.214844
3	1.0	1.0	0	6.56	0.816904
4	1.5	1.2247	0.1761	12.96	1.112605
5	2.0	1.4142	0.3010	18.71	1.272074
6	2.5	1.5811	0.3979	19.69	1.294246
7	3.0	1.7321	0.4771	20.35	1.308564
8	3.5	1.8708	0.5441	38.73	1.588047
9	4.0	2.0	0.6021	46.54	1.667826
10	4.5	2.1213	0.6532	59.58	1.7751
11	5.0	2.2361	0.6989	73.9	1.868644
12	5.5	2.3452	0.7404	80.6	1.906335
13	6.0	2.4495	0.7782	92.6	1.966611

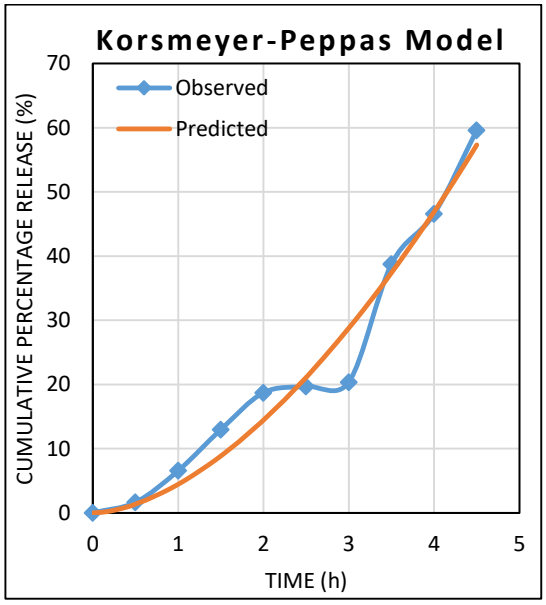
$R^2 = 0.9064$



$R^2 = 0.7947$



$R^2 = 0.6654$

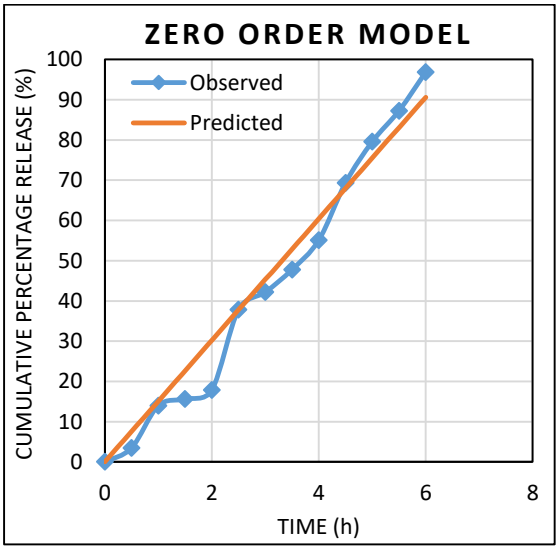


$R^2 = 0.9619$

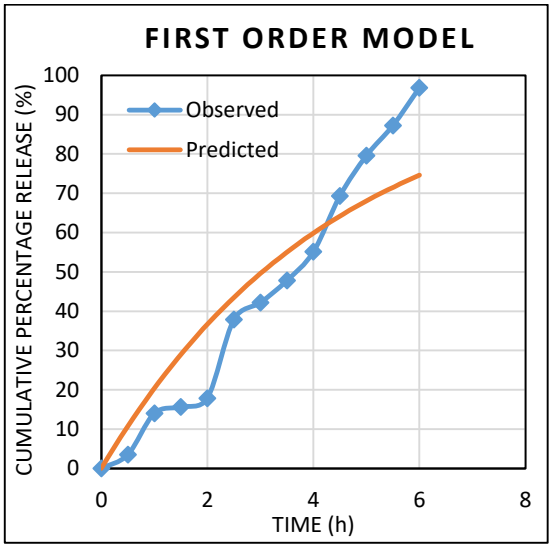
Fig-23: Kinetic models of drug release from F1

Table-13: Release kinetics data of F2 [BFR : Pectin – 4:4]

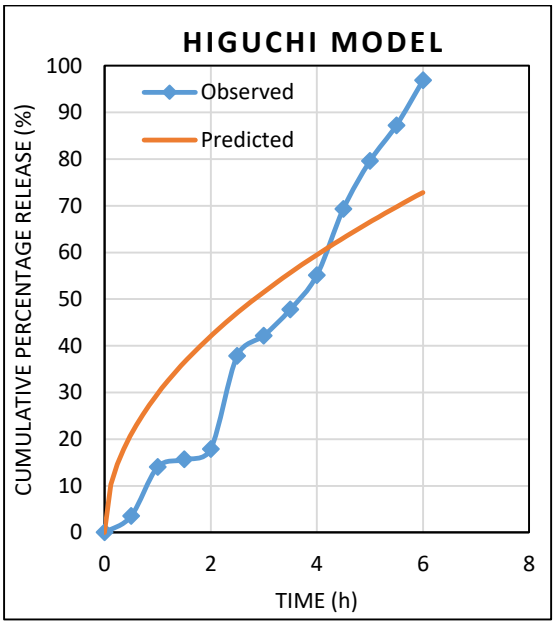
S.no	Time-t (h)	Square root of time - $t^{1/2}$	log t	Cumulative percentage of drug diffused-Q (%)	log Q
1	0	0	0	0	0
2	0.5	0.7071	-0.3010	3.49	0.542825
3	1.0	1.0	0	13.98	1.145507
4	1.5	1.2247	0.1761	15.63	1.193959
5	2.0	1.4142	0.3010	17.84	1.251395
6	2.5	1.5811	0.3979	37.84	1.577951
7	3.0	1.7321	0.4771	42.16	1.624901
8	3.5	1.8708	0.5441	47.78	1.679246
9	4.0	2.0	0.6021	55.09	1.741073
10	4.5	2.1213	0.6532	69.32	1.840859
11	5.0	2.2361	0.6989	79.56	1.900695
12	5.5	2.3452	0.7404	87.23	1.940666
13	6.0	2.4495	0.7782	96.84	1.986055



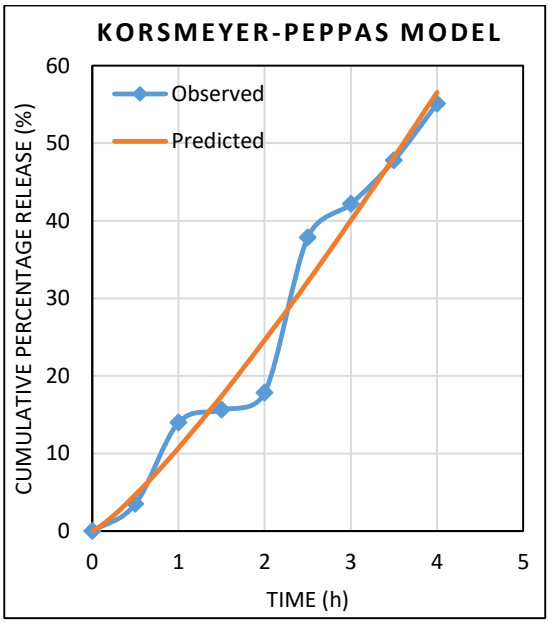
$R^2 = 0.9718$



$R^2 = 0.8670$



$R^2 = 0.7704$

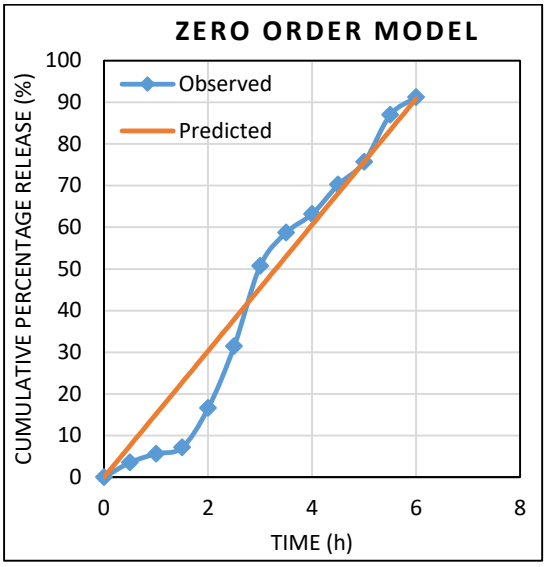


$R^2 = 0.9645$

Fig-24: Kinetic models of drug release from F2

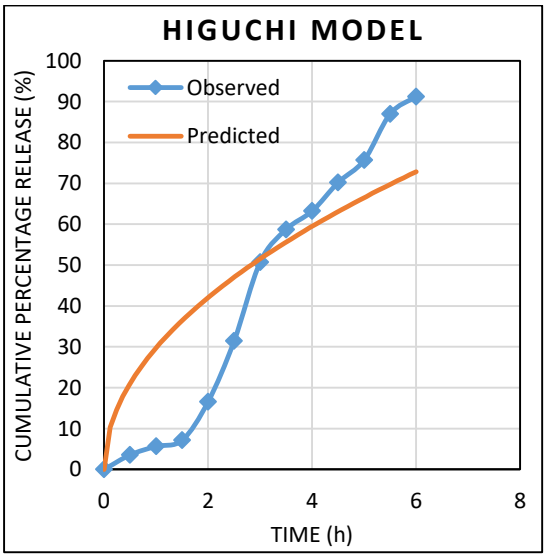
Table-14: Release kinetics data of F3 [BFR : Pectin – 5:3]

S.no	Time-t (h)	Square root of time - $t^{1/2}$	log t	Cumulative percentage of drug diffused-Q (%)	log Q
1	0	0	0	0	0
2	0.5	0.7071	-0.3010	3.57	0.552668
3	1.0	1.0	0	5.65	0.752048
4	1.5	1.2247	0.1761	7.143	0.853881
5	2.0	1.4142	0.3010	16.58	1.219585
6	2.5	1.5811	0.3979	31.43	1.497344
7	3.0	1.7321	0.4771	50.71	1.705094
8	3.5	1.8708	0.5441	58.73	1.76886
9	4.0	2.0	0.6021	63.21	1.800786
10	4.5	2.1213	0.6532	70.22	1.846461
11	5.0	2.2361	0.6989	75.68	1.878981
12	5.5	2.3452	0.7404	86.98	1.939419
13	6.0	2.4495	0.7782	91.23	1.960138

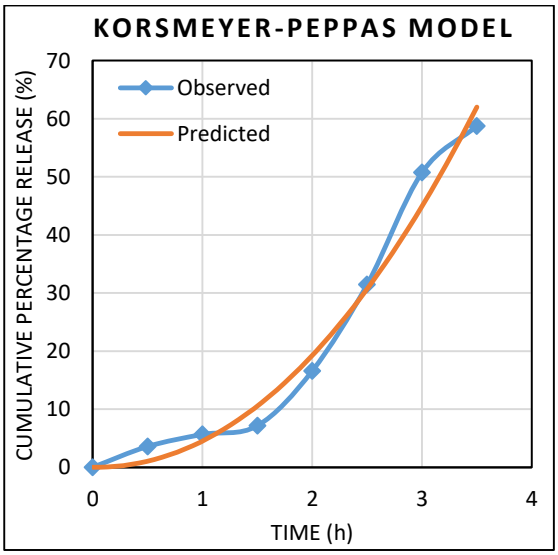


$R^2 = 0.9516$

$R^2 = 0.8590$



$R^2 = 0.7497$

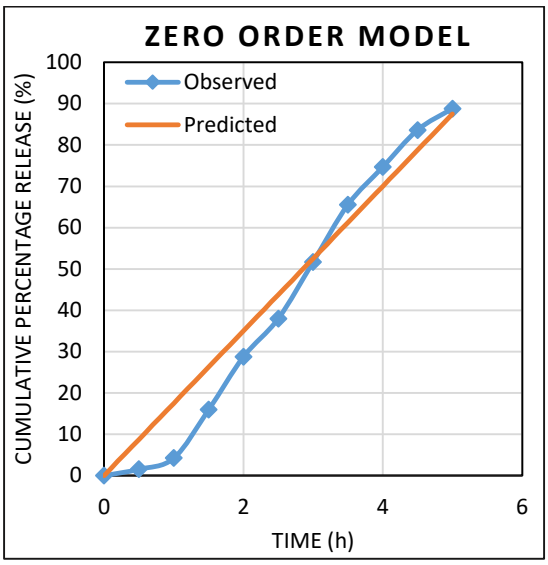


$R^2 = 0.9770$

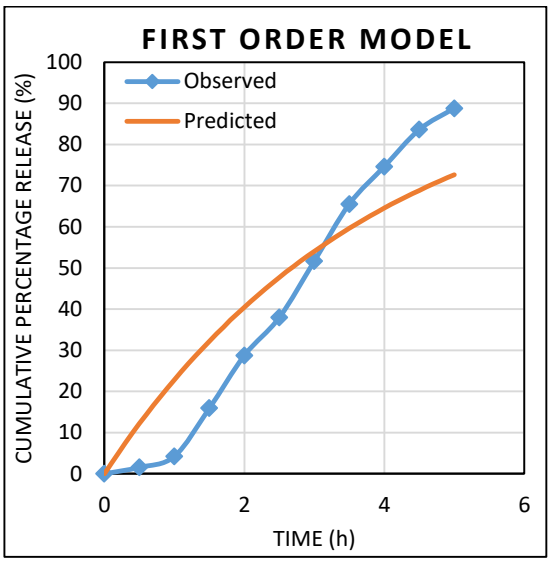
Fig-25: Kinetic models of drug release from F3

Table-15: Release kinetics data of F4 [BFR : SA – 4:2]

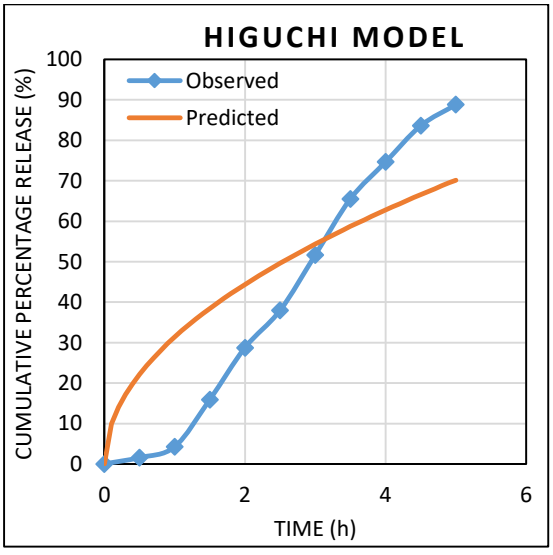
S.no	Time-t (h)	Square root of time - $t^{1/2}$	log t	Cumulative percentage of drug diffused-Q (%)	log Q
1	0	0	0	0	0
2	0.5	0.7071	-0.3010	1.56	0.19312
3	1.0	1.0	0	4.24	0.62737
4	1.5	1.2247	0.1761	15.93	1.20222
5	2.0	1.4142	0.3010	28.75	1.45864
6	2.5	1.5811	0.3979	37.98	1.57955
7	3.0	1.7321	0.4771	51.69	1.71341
8	3.5	1.8708	0.5441	65.54	1.81651
9	4.0	2.0	0.6021	74.65	1.87303
10	4.5	2.1213	0.6532	83.62	1.92231
11	5.0	2.2361	0.6989	88.82	1.94851
12	5.5	2.3452	0.7404	-	-
13	6.0	2.4495	0.7782	-	-



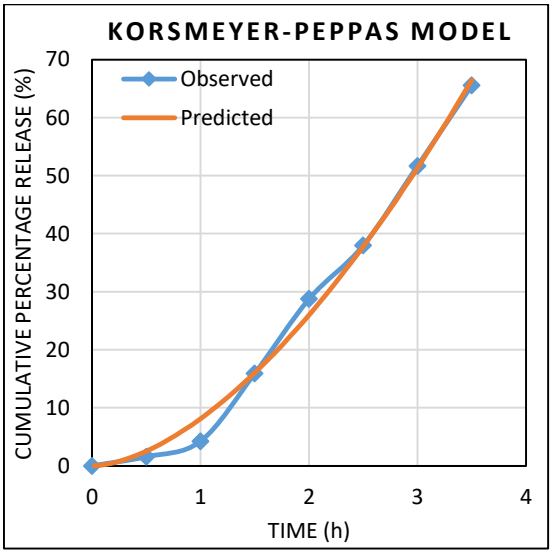
$R^2 = 0.9583$



$R^2 = 0.8608$



$R^2 = 0.7456$

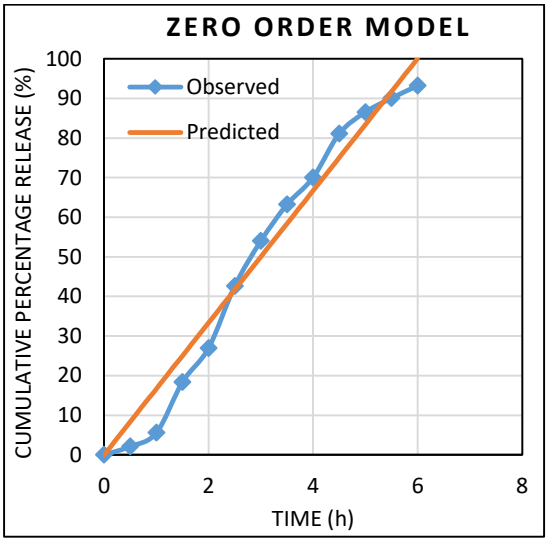


$R^2 = 0.9932$

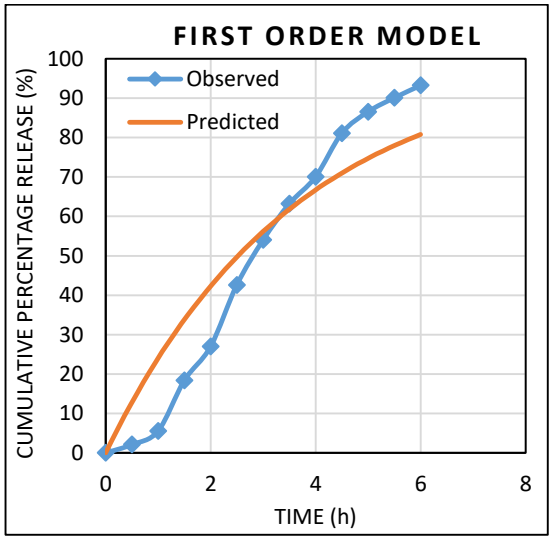
Fig-26: Kinetic models of drug release from F4

Table-16: Release kinetics data of F6 [BFR : SA – 4:3]

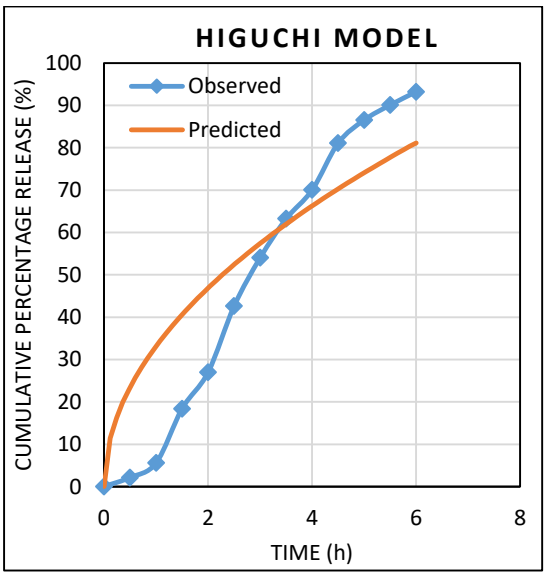
S.no	Time-t (h)	Square root of time - $t^{1/2}$	log t	Cumulative percentage of drug diffused-Q (%)	log Q
1	0	0	0	0	0
2	0.5	0.7071	-0.3010	2.12	0.32634
3	1.0	1.0	0	5.57	0.74586
4	1.5	1.2247	0.1761	18.39	1.26458
5	2.0	1.4142	0.3010	26.98	1.43104
6	2.5	1.5811	0.3979	42.61	1.62951
7	3.0	1.7321	0.4771	54.00	1.73239
8	3.5	1.8708	0.5441	63.21	1.80079
9	4.0	2.0	0.6021	70.02	1.84522
10	4.5	2.1213	0.6532	81.09	1.90897
11	5.0	2.2361	0.6989	86.54	1.93722
12	5.5	2.3452	0.7404	90.08	1.95463
13	6.0	2.4495	0.7782	93.21	1.96946



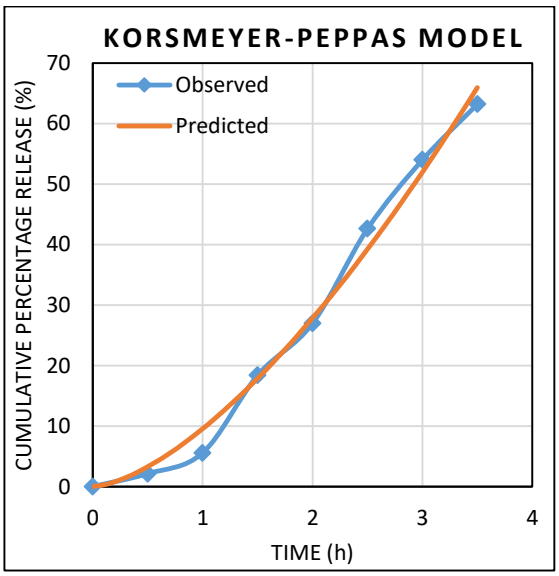
$R^2 = 0.9732$



$R^2 = 0.8952$



$R^2 = 0.8099$

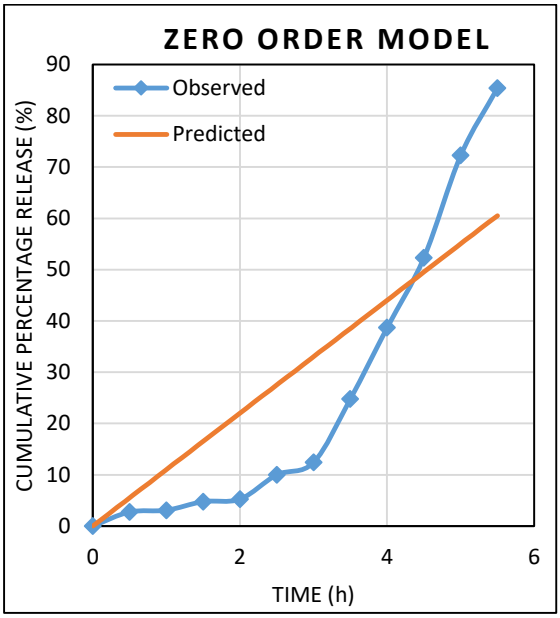


$R^2 = 0.9885$

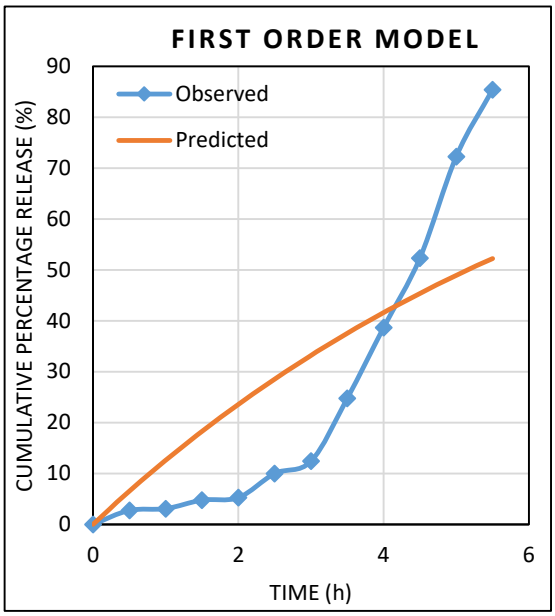
Fig-27: Kinetic models of drug release from F5

Table-17: Release kinetics data of F6 [BFR : SA – 4:3]

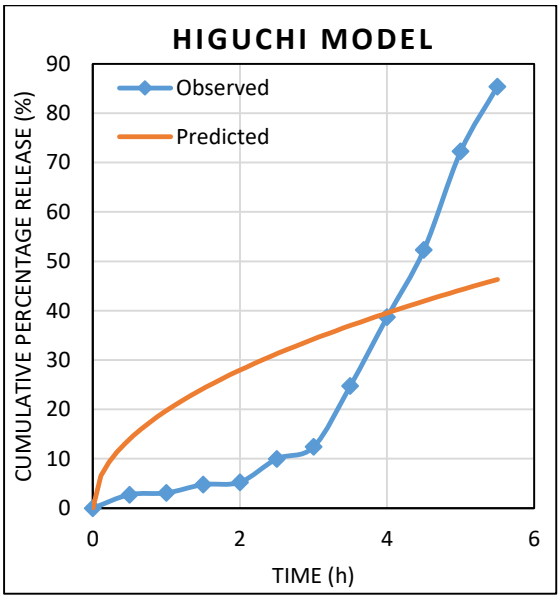
S.no	Time-t (h)	Square root of time - $t^{1/2}$	log t	Cumulative percentage of drug diffused-Q (%)	log Q
1	0	0	0	0	0
2	0.5	0.7071	-0.3010	2.73	0.43616
3	1.0	1.0	0	3.07	0.48714
4	1.5	1.2247	0.1761	4.76	0.67761
5	2.0	1.4142	0.3010	5.24	0.71933
6	2.5	1.5811	0.3979	9.98	0.99913
7	3.0	1.7321	0.4771	12.4	1.09342
8	3.5	1.8708	0.5441	24.77	1.39393
9	4.0	2.0	0.6021	38.68	1.58749
10	4.5	2.1213	0.6532	52.32	1.71867
11	5.0	2.2361	0.6989	72.26	1.8589
12	5.5	2.3452	0.7404	85.41	1.93151
13	6.0	2.4495	0.7782	-	-



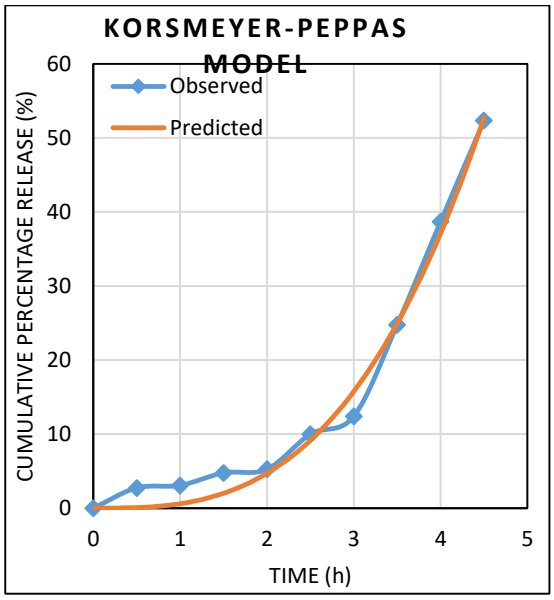
$R^2 = 0.7536$



$R^2 = 0.6599$



$R^2 = 0.4996$

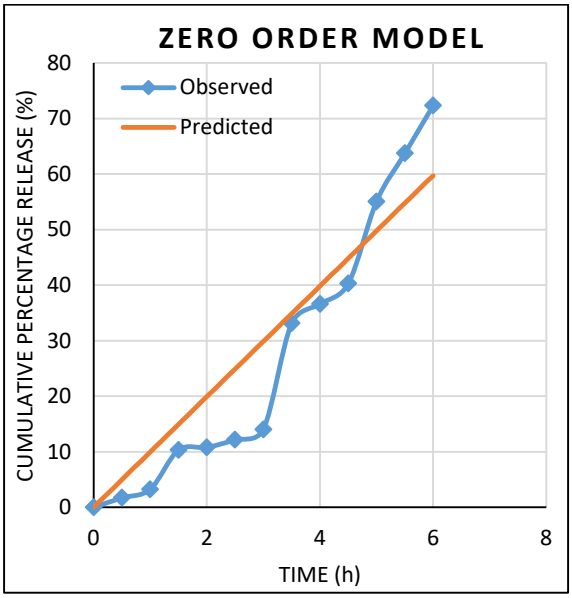


$R^2 = 0.9857$

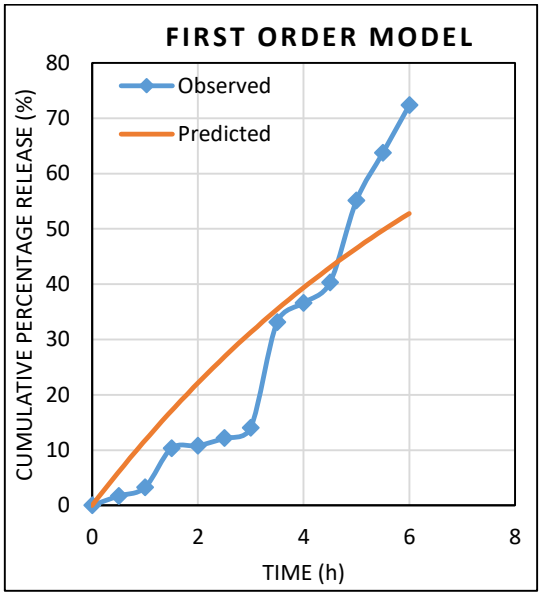
Fig-28: Kinetic models of drug release from F6

Table-18: Release kinetics data of F7 [BFR : PVA – 3:5]

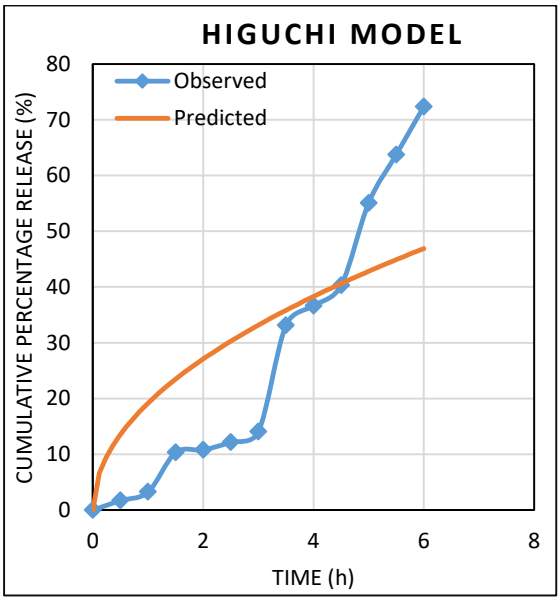
S.no	Time-t (h)	Square root of time - $t^{1/2}$	log t	Cumulative percentage of drug diffused-Q (%)	log Q
1	0	0	0	0	0
2	0.5	0.7071	-0.3010	1.72	0.23553
3	1.0	1.0	0	3.26	0.51322
4	1.5	1.2247	0.1761	10.32	1.01368
5	2.0	1.4142	0.3010	10.81	1.03383
6	2.5	1.5811	0.3979	12.17	1.08529
7	3.0	1.7321	0.4771	14.04	1.14737
8	3.5	1.8708	0.5441	33.14	1.52035
9	4.0	2.0	0.6021	36.61	1.5636
10	4.5	2.1213	0.6532	40.29	1.6052
11	5.0	2.2361	0.6989	55.09	1.74107
12	5.5	2.3452	0.7404	63.74	1.80441
13	6.0	2.4495	0.7782	72.35	1.85944



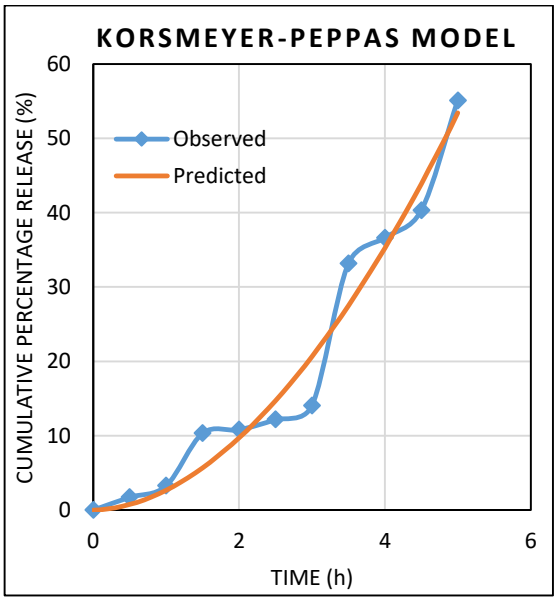
$R^2 = 0.8813$



$R^2 = 0.8028$



$R^2 = 0.6351$

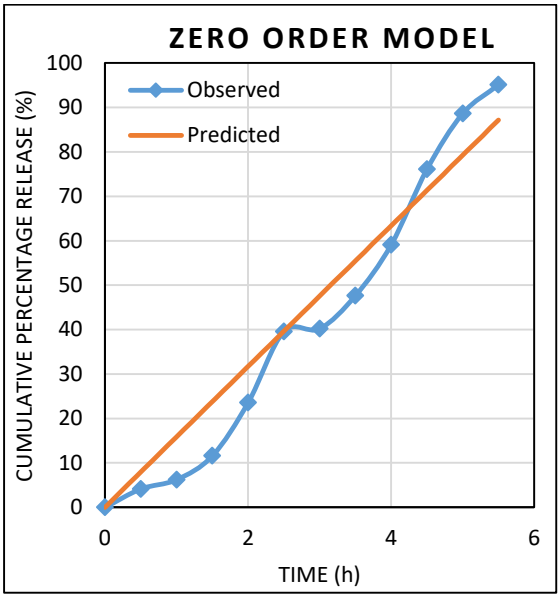


$R^2 = 0.9594$

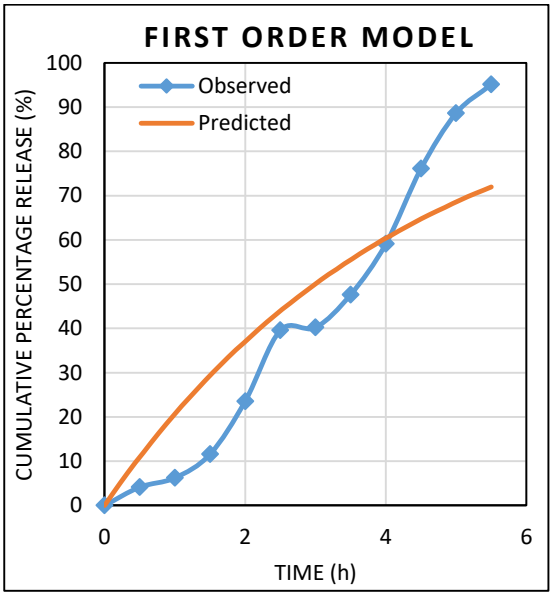
Fig-29: Kinetic models of drug release from F7

Table-19: Release kinetics data of F8 [BFR : PVA – 4:4]

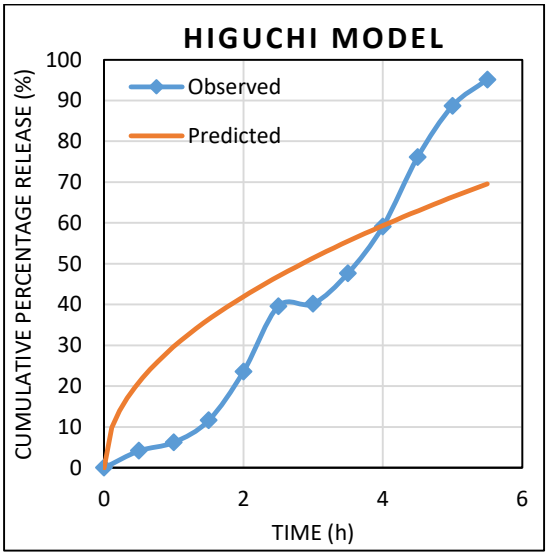
S.no	Time-t (h)	Square root of time - $t^{1/2}$	log t	Cumulative percentage of drug diffused-Q (%)	log Q
1	0	0	0	0	0
2	0.5	0.7071	-0.3010	4.15	0.61805
3	1.0	1.0	0	6.23	0.79449
4	1.5	1.2247	0.1761	11.59	1.06408
5	2.0	1.4142	0.3010	23.56	1.37218
6	2.5	1.5811	0.3979	39.58	1.59748
7	3.0	1.7321	0.4771	40.21	1.60433
8	3.5	1.8708	0.5441	47.65	1.67806
9	4.0	2.0	0.6021	59.10	1.77159
10	4.5	2.1213	0.6532	76.09	1.88133
11	5.0	2.2361	0.6989	88.64	1.94763
12	5.5	2.3452	0.7404	95.12	1.97827
13	6.0	2.4495	0.7782	-	



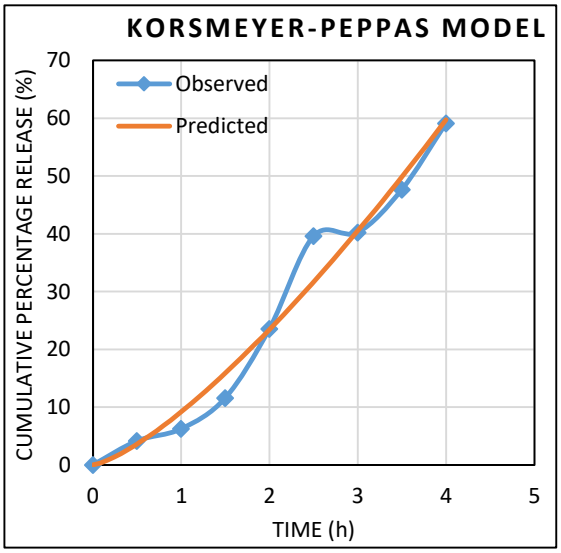
$R^2 = 0.9485$



$R^2 = 0.8365$



$R^2 = 0.7262$

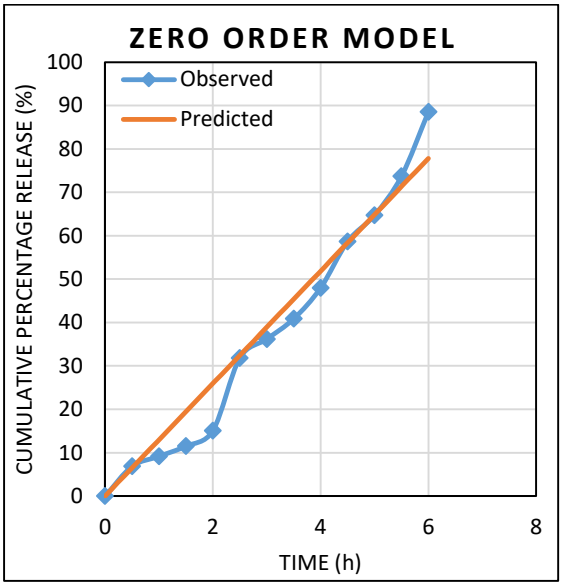


$R^2 = 0.9705$

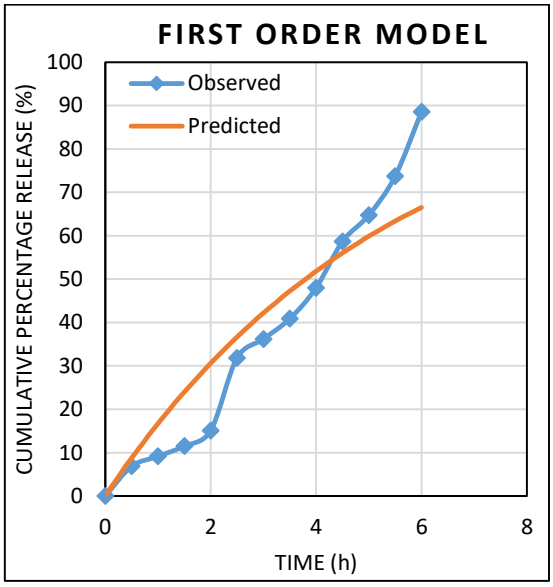
Fig-30: Kinetic models of drug release from F8

Table-20: Release kinetics data of F9 [BFR : PVA – 5:3]

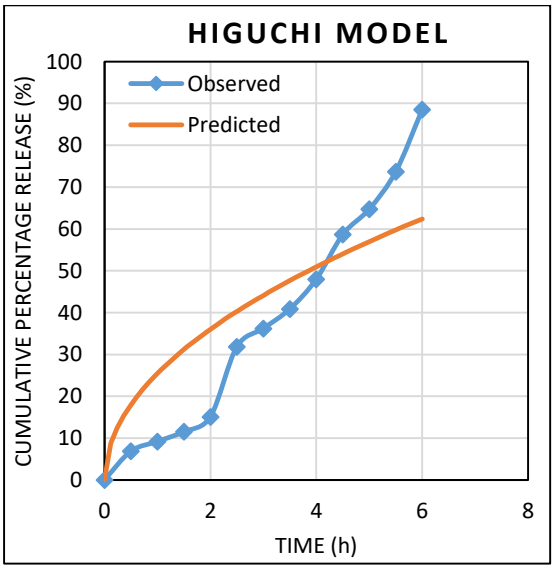
S.no	Time-t (h)	Square root of time - $t^{1/2}$	log t	Cumulative percentage of drug diffused-Q (%)	log Q
1	0	0	0	0	0
2	0.5	0.7071	-0.3010	6.86	0.83632
3	1.0	1.0	0	9.18	0.96284
4	1.5	1.2247	0.1761	11.52	1.06145
5	2.0	1.4142	0.3010	15.08	1.1784
6	2.5	1.5811	0.3979	31.83	1.50284
7	3.0	1.7321	0.4771	36.19	1.55859
8	3.5	1.8708	0.5441	40.86	1.6113
9	4.0	2.0	0.6021	47.98	1.68106
10	4.5	2.1213	0.6532	58.65	1.76827
11	5.0	2.2361	0.6989	64.72	1.81104
12	5.5	2.3452	0.7404	73.69	1.86741
13	6.0	2.4495	0.7782	88.51	1.94699



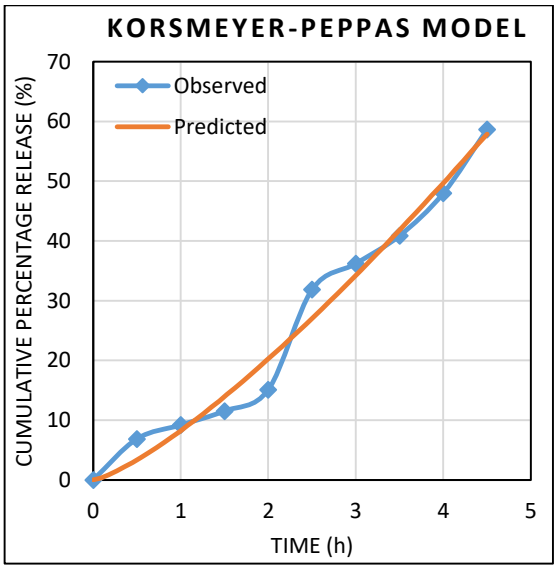
$R^2 = 0.9625$



$R^2 = 0.8760$



$R^2 = 0.7533$



$R^2 = 0.9757$

Fig-31: Kinetic models of drug release from F9

Correlation of coefficient values various kinetic models with respect to the in-vitro diffusion study were tabulated to determine the best-fit model and the mechanism of diffusion.

Table-21: Correlation of coefficient values various kinetic models

Formulation Code	Correlation coefficient value (R^2)			
	Zero order kinetic Model	First order kinetic Model	Higuchi's Model	Korsmeyer-Peppas Model
F1	0.9064	0.7947	0.6654	0.9619
F2	0.9718	0.8670	0.7704	0.9645
F3	0. 9516	0.8590	0.7497	0.9770
F4	0.9583	0.8608	0.7456	0.9932
F5	0. 9732	0.8952	0.8099	0.9885
F6	0. 7536	0.6599	0.4996	0.9857
F7	0.8813	0.8028	0.6351	0.9594
F8	0.9485	0.8365	0.7262	0.9705
F9	0.9625	0.8760	0.7533	0.9757
Average	0.9381	0.8279	0.7061	0.9752
Standard deviation	0.04	0.07	0.09	0.01

Inference

In-vitro permeation studies revealed that the formulation F7 (BFR : PVA- 3:5) exhibits a sustained release of more than 6 hrs and hence PVA is a suitable combination for BFR for a sustained release drug delivery.

The release kinetic modelling shows that the formulated Metadoxine buccal patches undergo **zero order kinetic release**, since the correlation coefficient values corresponding to zero order model of all the formulations are comparatively higher and closer to 1.0 (averaging at 0.9381±0.04) than First order and Higuchi models.

The Korsmeyer-Peppas modelling helped to determine the release mechanism of the buccal patch formulations as **'non-Fickian mechanism**' (according to Table-4 &20), since the average 'n' exponent value is 0.9752 ± 0.01.

SUMMARY

Natural polymers are trending as a reliable alternative for synthetic and semi synthetic polymers, in the development of a large number of novel drug delivery systems. One such new alternative to be used as a mucoadhesive polymer- **B. flabellifer Fruit resin**, especially for buccal drug delivery was introduced.

The novel polymer, in combination with two other natural polymers (Pectin & Sodium alginate) and one synthetic polymer (PVA), was used to formulate a buccal drug delivery system containing **Metadoxine**. This drug was chosen due to its low half-life (maximum of 60 min) and attempt was made to reduce its dose by sustaining its release. Also alcoholism is a serious social and health issue affecting a significant amount of world population and hence a therapeutic alternative to cure alcoholism is a need of the hour.

Compatibility studies carried out with the help of FT-IR spectrometer indicated that there are no chemical interactions between the drug and the polymers used, including BFR. The calibration graph of Metadoxine was obtained by a validated UV spectrophotometric method at λ_{max} of 324 nm.

BFR was extracted from ripened palm fruit; stored and used for formulating 9 formulations in the ratios BFR : Pectin - 3:5, 4:4, 5:3 / BFR : SA – 4:2, 4:3, 4:4 and BFR : PVA - 3:5, 4:4, 5:3 respectively (the numbers in the ratios indicate the polymer concentration in percentage). A backing membrane of 4% PVA was also coated over one side of all formulations.

Physico-chemical properties such as thickness, weight variation, folding endurance, swelling index, surface pH, drug content and bioadhesion strength were evaluated appropriately and, the results were tabulated and compared. In-vitro diffusion study was also performed to examine the release pattern of the formulations, which was extended to determine the kinetics and mechanism of the release.

CONCLUSION

Metadoxine buccal patches were formulated and evaluated successfully by solvent casting method; following standard operating procedures. The evaluation tests revealed that *B. flabellifer* is a suitable polymer for developing a sustained release buccal drug delivery system. Among the developed buccal patches, the formulation F7 with a polymer combination of 3% w/v BFR and 5% w/v PVA seems to be an optimized formulation, since it exhibits better folding endurance, uniformity of drug content, and sustained release of drug. Therefore, Metadoxine which exhibits lower elimination half-life can be incorporated in buccal drug delivery systems, in order decrease the dose frequency and thereby decreasing the possibility of dose dumping.

It also should be noted that, concentration of BFR is directly proportional to the bioadhesion strength and hence BFR justifies its selection as a novel mucoadhesive polymer of natural origin.

FUTURE PLAN & EXTENSION OF THE WORK

This study proves that resin obtained from fruits of *B. flabellifer* has the potential to be used as formidable natural polymer. Hence it can also be used as

- ✓ thickening agent or viscosity modifier
- ✓ binding agent (when solubilized at low concentrations)
- ✓ gelling agent

If further attempts are made, it can be used alone as a film-forming polymer with the help of varying plasticizers [23].

In another aspect, this polymer can also be incorporated with fast disintegrating agents and developed into fast dissolving films.

It is also evident that the plant possesses the same constituents elsewhere among its parts. One such variation in the source of this polymer can be unripe fruits of *B. flabellifer* [18]. But the difference is, it can be obtained as a coarse powder than a resin, if dried properly.

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ANNEXURE

भारत सरकार
GOVERNMENT OF INDIA
पर्यावरण, वन और जलवायु परिवर्तन मंत्रालय
MINISTRY OF ENVIRONMENT, FOREST & CLIMATE CHANGE
भारतीय वनस्पति सर्वेक्षण
BOTANICAL SURVEY OF INDIA

दक्षिणी क्षेत्रीय केन्द्र / Southern Regional Centre
टी.एन.ए.यू कैम्पस / T.N.A.U. Campus
लाउली रोड / Lawley Road
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bsisc@rediffmail.com

सं. भा.व.स./द.क्षे.के./No.: BSI/SRC/5/23/2017/Tech. 218 दिनांक/Date: 2nd May 2017

सेवा में / To

Mr. V. Hariharshan
II Year M. Pharmacy
Department of Pharmaceutics
College of Pharmacy – Sri Ramakrishna Institute of Paramedical Sciences
Coimbatore – 641 044

महोदय/Sir,

The plant specimen brought by you for authentication is identified as *Borassus flabellifer* L. - ARECACEAE. The identified specimen is returned herewith for preservation in their College/ Department/ Institution Herbarium.

धन्यवाद/Thanking you,

भवदीय/Yours faithfully,

(डॉ सी मुरुगन/Dr. C. Murugan)
वैज्ञानिक 'डी' एवं कार्यालय अध्यक्ष /
Scientist 'D' & Head of Office

वैज्ञानिक 'डी' एवंकार्यालयअध्यक्ष
SCIENTIST 'D' & Head of Office
भारतीय वनस्पति सर्वेक्षण
Botanical Survey of India
दक्षिणी क्षेत्रीय केन्द्र
Southern Regional Centre
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